Methodology for Deriving Ambient Water Quality Criteria for the Protection of Human Health (2000)

Chapter 5

TABLES AND FIGURES

5. BIOACCUMULATION

5.1 INTRODUCTION

Aquatic organisms can accumulate certain chemicals in their bodies when exposed to these chemicals through water, their diet, and other sources. This process is called bioaccumulation. The magnitude of bioaccumulation by aquatic organisms varies widely depending on the chemical but can be extremely high for some highly persistent and hydrophobic chemicals. For such highly bioaccumulative chemicals, concentrations in aquatic organisms may pose unacceptable human health risks from fish and shellfish consumption even when concentrations in water are too low to cause unacceptable health risks from drinking water consumption alone. These chemicals may also biomagnify in aquatic food webs, a process whereby chemical concentrations increase in aquatic organisms of each successive trophic level due to increasing dietary exposures (e.g., increasing concentrations from algae, to zooplankton, to forage fish, to predatory fish).

In order to prevent harmful exposures to waterborne chemicals through the consumption of contaminated fish and shellfish, national 304(a) water quality criteria for the protection of human health must address the process of chemical bioaccumulation in aquatic organisms. For deriving national 304(a) criteria to protect human health, EPA accounts for potential bioaccumulation of chemicals in fish and shellfish through the use of national bioaccumulation factors (BAFs). A national BAF is a ratio (in L/kg) that relates the concentration of a chemical in water to its expected concentration in commonly consumed aquatic organisms in a specified trophic level. An illustration of how national BAFs are used in the derivation of 304(a) criteria for carcinogens using linear low-dose extrapolation is shown in the following equation:

$$AWQC = RSD \bullet \left(\frac{BW}{DI + \sum_{i=2}^{4} (FI_i \bullet BAF_i)} \right) \qquad \text{(Equation 5-1)}$$

where:

RSD	=	Risk specific dose (mg/kg-day)
BW	=	Human body weight (kg)
DI	=	Drinking water intake (L/day)
FI_i	=	Fish intake at trophic level I, where I=2, 3, and 4;
BAF_i	=	National bioaccumulation factor at trophic level I, where I=2, 3, and 4

The purpose of this chapter is to present EPA's recommended methodology for deriving national bioaccumulation factors for setting national 304(a) water quality criteria to protect human health. A detailed scientific basis of the recommended national BAF methodology is provided in the Bioaccumulation TSD. While the methodology detailed in this chapter is

intended to be used by EPA for deriving national BAFs, EPA encourages States and authorized Tribes to derive BAFs that are specific to certain regions or waterbodies, where appropriate. Guidance to States and authorized Tribes for deriving site-specific BAFs is provided in the Biaccumulation TSD.

5.1.1 Important Bioaccumulation and Bioconcentration Concepts

Several attributes of the bioaccumulation process are important to understand when deriving national BAFs for use in setting national 304(a) criteria. First, the term "bioaccumulation" refers to the uptake and retention of a chemical by an aquatic organism from all surrounding media (e.g., water, food, sediment). The term "bioconcentration" refers to the uptake and retention of a chemical by an aquatic organism from water only. For some chemicals (particularly those that are highly persistent and hydrophobic), the magnitude of bioaccumulation by aquatic organisms can be substantially greater than the magnitude of bioconcentration. Thus, an assessment of bioconcentration alone would underestimate the extent of accumulation in aquatic biota for these chemicals. Accordingly, EPA's guidelines presented in this chapter emphasize the measurement of chemical bioaccumulation by aquatic organisms, whereas EPA's 1980 Methodology emphasized the measurement of bioconcentration.

Another noteworthy aspect of bioaccumulation process is the issue of steady-state conditions. Specifically, both bioaccumulation and bioconcentration can be viewed simply as the result of competing rates of chemical uptake and depuration (chemical loss) by an aquatic organism. The rates of chemical uptake and depuration can be affected by various factors including the properties of the chemical, the physiology of the organism in question, water quality and other environmental conditions, ecological characteristics of the waterbody (e.g., food web structure), and the concentration and loadings history of the chemical. When the rates of chemical uptake and depuration are equal, tissue concentrations remain constant over time and the distribution of the chemical between the organism and its source(s) is said to be at steady-state. For constant chemical exposures and other conditions, the steady-state concentration in the organism represents the highest accumulation potential of the chemical in that organism under those conditions. The time required for a chemical to achieve steady state has been shown to vary according to the properties of the chemical and other factors. For example, some highly hydrophobic chemicals can require long periods of time to reach steady state between environmental compartments (e.g., many months), while highly hydrophilic chemicals usually reach steady-state relatively quickly (e.g., hours to days).

Since national 304(a) criteria for the protection of human health are typically designed to protect humans from harmful lifetime or long-term exposures to waterborne contaminants, the assessment of bioaccumulation that equals or approximates steady-state accumulation is one of the principles underlying the derivation of national BAFs. For some chemicals that require relatively long periods of time to reach steady-state in tissues of aquatic organisms, changes in water column concentrations may occur on a much more rapid time scale compared to the corresponding changes in tissue concentrations. Thus, if the system departs substantially from steady-state conditions and water concentrations are not averaged over a sufficient time period,

the ratio of the tissue concentration to a water concentration may have little resemblance to the steady-state ratio and have little predictive value of long-term bioaccumulation potential. Therefore, BAF measurements should be based on water column concentrations which are averaged over a sufficient period of time (e.g., a duration comparable to the time required for the chemical to reach steady-state). In addition, BAF measurements should be based on adequate spatial averaging of both tissue and water column concentrations for use in deriving 304(a) criteria for the protection of human health.

For this reason, a BAF is defined in this Methodology as representing the ratio (in L/kg-tissue) of a concentration of a chemical in tissue to its concentration in the surrounding water in situations where the organism and its food are exposed and the ratio does not change substantially over time (i.e., the ratio which reflects bioaccumulation at or near steady-state). A bioconcentration factor (BCF) is the ratio (in L/kg-tissue) of the concentration of a substance in tissue of an aquatic organism to its concentration in the ambient water, in situations where the organism is exposed through the water only and the ratio does not change substantially over time.

5.1.2 Goal of the National BAF

The goal of EPA's national BAF is to represent the long-term, average bioaccumulation potential of a chemical in edible tissues of aquatic organisms that are commonly consumed by humans throughout the United States. National BAFs are not intended to reflect fluctuations in bioaccumulation over short time periods (e.g., a few days) because 304(a) human health criteria are generally designed to protect humans from long-term exposures to waterborne chemicals. National BAFs are also intended to account for some major chemical, biological, and ecological attributes that can affect bioaccumulation in bodies of water across the United States. For example, separate procedures are provided for deriving national BAFs depending on the type of chemical (i.e., nonionic organic, ionic organic, inorganic and organometallic). In addition, EPA's national BAFs are derived separately for each trophic level to account for potential biomagnification of some chemicals in aquatic food webs and broad physiological differences between trophic levels that may influence bioaccumulation. Because lipid content of aquatic organisms and the amount of organic carbon in the water column have been shown to affect bioaccumulation of nonionic organic chemicals, EPA's national BAFs are adjusted to reflect the lipid content of commonly consumed fish and shellfish and the freely dissolved fraction of the chemical in ambient water for these chemicals.

5.1.3 Changes to the 1980 Methodology

Numerous scientific advances have occurred in the area of bioaccumulation since the publication of the 1980 Methodology for deriving AWQC for the protection of human health (USEPA, 1980). These advances have significantly increased our ability to assess and predict the bioaccumulation of chemicals in aquatic biota. As a result, EPA has revised the bioaccumulation portion of the 1980 Methodology to reflect the current state of the science and to improve accuracy in assessing bioaccumulation for setting 304(a) criteria for the protection of

human health. The changes contained in the bioaccumulation portion of the 2000 Human Health Methodology are mostly designed to:

- Improve the ability to incorporate chemical exposure from sediments and aquatic food webs in assessing bioaccumulation potential,

- Expand the ability to account for site-specific factors which affect bioaccumulation, and

- Incorporate new data and assessment tools into the bioaccumulation assessment process.

A summary of the key changes that have been incorporated into the bioaccumulation portion of the 2000 Human Health Methodology and appropriate comparisons to the1980 Methodology are provided below.

5.1.3.1 Overall Approach

The 1980 Methodology for deriving 304(a) criteria for the protection of human health emphasized the assessment of bioconcentration (uptake from water only) through the use of the BCF. Based on the 1980 Methodology, measured BCFs were usually determined from laboratory data unless field data demonstrated consistently higher or lower accumulation compared with laboratory data. In these cases, "field BCFs" (currently termed field-measured BAFs) were recommended for use. For lipophilic chemicals where lab or field-measured data were unavailable, EPA recommended predicting BCFs from the octanol-water partition coefficient and the following equation from Veith et al. (1979): "log BCF = (0.85 log K_{ow}) - 0.70".

The 2000 Human Health Methodology revisions contained in this chapter emphasize the measurement of bioaccumulation (uptake from water, sediment, and diet) through the use of the BAF. Consistent with the 1980 Methodology, measured data are preferred over predictive approaches for determining the BAF (i.e., field-measured BAFs are generally preferred over predicted BAFs). However, the 2000 Human Health Methodology contains additional methods for deriving a national BAF that were not available in 1980. The preference for using the BAF methods also differs depending on the type and properties of the chemical. For example, the BAF derivation procedure differs for each of three broadly defined chemical categories: (1) nonionic organic, (2) ionic organic, and (3) inorganic and organometallic chemicals. Furthermore, within the category of nonionic organic chemicals, different procedures are used to derive the BAF depending on a chemicals' hydrophobicity and extent of chemical metabolism that would be expected to occur in aquatic biota.

5.1.3.2 Lipid Normalization

In the 1980 Methodology, BCFs for lipophilic chemicals were normalized by the lipid fraction in the tissue of fish and shellfish used to determine the BCF. Lipid normalization enabled BCFs to be averaged across tissues and organisms. Once the average lipid-normalized

BCF was determined, it was adjusted by the consumption-weighted lipid content of commonly consumed aquatic organisms in the United States to obtain an overall consumption-weighted BCF. A similar procedure has been retained in the 2000 Human Health Methodology, whereby BAFs for nonionic organic chemicals are lipid normalized and adjusted by the consumption-weighted lipid content of commonly consumed organisms to obtain a BAF for criteria calculations. However, the 2000 Human Health Methodology uses more up-to-date lipid data and consumption data for deriving the consumption-weighted BAFs.

5.1.3.3 Bioavailability

Bioconcentration factors derived according to the 1980 Methodology were based on the total concentration of the chemical in water, for both lipophilic and nonlipophilic chemicals. In the 2000 Human Health Methodology, BAFs for nonionic organic chemicals are derived using the most bioavailable fraction (i.e., the freely dissolved fraction) to account for the influence of particulate and dissolved organic carbon on a chemical's bioavailability. Such BAFs are then adjusted to reflect the expected bioavailability at the sites of interest (i.e., by adjusting for organic carbon concentrations at the sites of interest). Procedures for accounting for the effect of organic carbon on bioaccumulation were published previously by EPA under the Great Lakes Water Quality Initiative (GLWQI or GLI) rulemaking (USEPA, 1995a,b). Bioavailability is also considered in developing BAFs for the other chemical classes defined in the 2000 Human Health Methodology (e.g., ionic organics, inorganics/organometallics) but is done so on a chemical-by-chemical basis.

5.1.3.4 Trophic Level Considerations

In the 1980 Methodology, BCFs were determined and used for criteria derivation without explicit regard to the trophic level of the aquatic organism (e.g., benthic filter feeder, forage fish, predatory fish). Over the past two decades, much information has been assembled which demonstrates that an organism's trophic position in the aquatic food web can have an important effect on the magnitude of bioaccumulation of certain chemicals. In order to account for the variation in bioaccumulation that is due to trophic position of the organism, the 2000 Human Health Methodology recommends that BAFs be determined and applied on a trophic level-specific basis.

5.1.3.5 Site-Specific Adjustments

The 1980 Methodology contained little guidance for making adjustments to the national BCFs to reflect site- or region-specific conditions. The 2000 Human Health Methodology has greatly expanded the guidance to States and authorized Tribes for making adjustments to national BAFs to reflect local conditions. This guidance is contained in the Bioaccumulation TSD. In the Bioaccumulation TSD, guidance and data are provided for adjusting national BAFs to reflect the lipid content in locally consumed aquatic biota and the organic carbon content in the waterbodies of concern. This guidance also allows the use of appropriate bioaccumulation models for deriving site-specific BAFs. EPA also plans to publish detailed guidance on designing and conducting field

bioaccumulation studies for measuring BAFs and biota-sediment accumulation factors (BSAFs). In general, EPA encourages States and authorized Tribes to make site-specific modifications to EPA's national BAFs provided such adjustments are scientifically defensible and adequately protect the designated use of the waterbody.

While the aforementioned revisions are new to EPA's Methodology for deriving national 304(a) criteria for the protection of human health, many of these refinements have been incorporated in prior Agency guidance and regulations. For example, the use of food chain multipliers to account for the biomagnification of nonionic organic chemicals in aquatic food webs when measured data are unavailable was introduced by EPA in three documents*: Technical Support Document for Water Quality-Based Toxics Control* (USEPA, 1991), a draft document entitled *Assessment and Control of Bioconcentratable Contaminants in Surface Waters* (USEPA, 1993), and in the *Great Lakes Water Quality Initiative* (GLI) (USEPA, 1995b). Similarly, procedures for predicting BAFs using BSAFsand incorporating the effect of organic carbon on bioavailability were used to derive water quality criteria under the GLI.

5.1.4 Organization of This Section

The methodology for deriving national BAFs for use in deriving National 304(a) Human Health AWQC is provided in the following sections. Important terms used throughout this chapter are defined in Section 5.2. Section 5.3 provides an overview of the BAF derivation guidelines. Detailed procedures for deriving national BAFs are provided in Section 5.4 for nonionic organic chemicals, in Section 5.5 for ionic organic chemicals, and in Section 5.6 for inorganics and organometallic chemicals. Literature cited is provided in Section 5.7.

5.2 DEFINITIONS

The following terms and definitions are used throughout this chapter.

Bioaccumulation. The net accumulation of a substance by an organism as a result of uptake from all environmental sources.

Bioconcentration. The net accumulation of a substance by an aquatic organism as a result of uptake directly from the ambient water, through gill membranes or other external body surfaces.

Bioaccumulation Factor (BAF). The ratio (in L/kg-tissue) of the concentration of a substance in tissue to its concentration in the ambient water, in situations where both the organism and its food are exposed and the ratio does not change substantially over time. The BAF is calculated as:

$$BAF = \frac{C_t}{C_w} \qquad \text{(Equation 5-2)}$$

where:

C_t = Concentration of the chemical in the specified wet tissue
C_w = Concentration of chemical in water

Bioconcentration Factor (BCF). The ratio (in L/kg-tissue) of the concentration of a substance in tissue of an aquatic organism to its concentration in the ambient water, in situations where the organism is exposed through the water only and the ratio does not change substantially over time. The BCF is calculated as:

$$BCF = \frac{C_t}{C_w} \qquad \text{(Equation 5-3)}$$

where:

C_t = Concentration of the chemical in the specified wet tissue
C_w = Concentration of chemical in water

Baseline BAF (BAF_R^{fd}). For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), a BAF (in L/kg-lipid) that is based on the concentration of freely dissolved chemical in the ambient water and the lipid normalized concentration in tissue.

Baseline BCF (BCF_R^{fd}). For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), a BCF (in L/kg-lipid) that is based on the concentration of freely dissolved chemical in the ambient water and the lipid normalized concentration in tissue.

Biomagnification. The increase in tissue concentration of a chemical in organisms at successive trophic levels through a series of predator-prey associations, primarily through the mechanism of dietary accumulation.

Biomagnification Factor (BMF). The ratio (unitless) of the tissue concentration of a chemical in a predator at a particular trophic level to the tissue concentration in its prey at the next lower trophic level for a given waterbody and chemical exposure. For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), a BMF can be calculated using lipid-normalized concentrations in the tissue of organisms at two successive trophic levels as:

$$BMF_{(TL,\ n)} = \frac{C_{\ell\ (TL,\ n)}}{C_{\ell\ (TL,\ n-1)}} \qquad \text{(Equation 5-4)}$$

where:

$C_{R\,(TL,\,n)}$ = Lipid-normalized concentration in appropriate tissue of predator organism at a given trophic level (TL "n")

$C_{R\,(TL,\,n-1)}$ = Lipid-normalized concentration in appropriate tissue of prey organism at the next lower trophic level from the predator (TL "n-1")

For inorganic, organometallic, and certain ionic organic chemicals where lipid and organic carbon partitioning does not apply, a BMF can be calculated using chemical concentrations in the tissue of organisms at two successive trophic levels as:

$$BMF_{(TL,\,n)} = \frac{C_{t\,(TL,\,n)}}{C_{t\,(TL,\,n-1)}} \qquad \text{(Equation 5-5)}$$

where:

$C_{t\,(TL,\,n)}$ = Concentration in appropriate tissue of predator organism at trophic level "n" (may be either wet weight or dry weight concentration so long as both the predator and prey concentrations are expressed in the same manner)

$C_{t\,(TL,\,n-1)}$ = Concentration in appropriate tissue of prey organism at the next lower trophic level from the predator (may be either wet weight or dry weight concentration so long as both the predator and prey concentrations are expressed in the same manner)

Biota-Sediment Accumulation Factor (BSAF). For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), the ratio of the lipid-normalized concentration of a substance in tissue of an aquatic organism to its organic carbon-normalized concentration in surface sediment (expressed as kg of sediment organic carbon per kg of lipid), in situations where the ratio does not change substantially over time, both the organism and its food are exposed, and the surface sediment is representative of average surface sediment in the vicinity of the organism. The BSAF is defined as:

$$BSAF = \frac{C_{\ell}}{C_{soc}} \qquad \text{(Equation 5-6)}$$

where:

C_R = The lipid-normalized concentration of the chemical in tissues of the biota (µg/g lipid)

C_{soc} = The organic carbon-normalized concentration of the chemical in the surface sediment (µg/g sediment organic carbon)

Depuration. The loss of a substance from an organism as a result of any active or passive process.

Food Chain Multiplier (FCM). For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), the ratio of a baseline BAF_R^{fd} for an organism of a particular trophic level to the baseline BCF_R^{fd} (usually determined for organisms in trophic level one). For inorganic, organometallic, and certain ionic organic chemicals where lipid and organic carbon partitioning does not apply, a FCM is based on total (wet or dry weight) concentrations of the chemical in tissue.

Freely Dissolved Concentration. For nonionic organic chemicals, the concentration of the chemical that is dissolved in ambient water, excluding the portion sorbed onto particulate or dissolved organic carbon. The freely dissolved concentration is considered to represent the most bioavailable form of an organic chemical in water and, thus, is the form that best predicts bioaccumulation. The freely dissolved concentration can be determined as:

$$C_w^{fd} = (C_w^t) \cdot (f_{fd}) \qquad \text{(Equation 5-7)}$$

where:

C_w^{fd} = Freely dissolved concentration of the organic chemical in ambient water
C_w^t = Total concentration of the organic chemical in ambient water
f_{fd} = Fraction of the total chemical in ambient water that is freely dissolved

Hydrophilic. A term that refers to the extent to which a chemical is attracted to partitioning into the water phase. Hydrophilic organic chemicals have a greater tendency to partition into polar phases (e.g., water) compared to chemicals of hydrophobic chemicals.

Hydrophobic. A term that refers to the extent to which a chemical avoids partitioning into the water phase. Highly hydrophobic organic chemicals have a greater tendency to partition into nonpolar phases (e.g., lipid, organic carbon) compared with chemicals of lower hydrophobicity.

Lipid-normalized Concentration (C_l). The total concentration of a contaminant in a tissue or whole organism divided by the lipid fraction in that tissue or whole organism. The lipid-normalized concentration can be calculated as:

$$C_l = \frac{C_t}{f_l} \qquad \text{(Equation 5-8)}$$

where:

C_t = Concentration of the chemical in the wet tissue (either whole organism or specified tissue)

f_R = Fraction lipid content in the organism or specified tissue

Octanol-water Partition Coefficient (K_{ow}). The ratio of the concentration of a substance in the n-octanol phase to its concentration in the aqueous phase in an equilibrated two-phase octanol-water system. For log K_{ow}, the log of the octanol-water partition coefficient is a base 10 logarithm.

Organic Carbon-normalized Concentration (C_{soc}). For sediments, the total concentration of a contaminant in sediment divided by the fraction of organic carbon in sediment. The organic carbon-normalized concentration can be calculated as:

$$C_{soc} = \frac{C_s}{f_{oc}} \qquad \text{(Equation 5-9)}$$

where:

C_s = Concentration of chemical in sediment

f_{oc} = Fraction organic carbon in sediment

Uptake. Acquisition by an organism of a substance from the environment as a result of any active or passive process.

5.3 FRAMEWORK FOR DETERMINING NATIONAL BIOACCUMULATION FACTORS

5.3.1 Four Different Methods

Bioaccumulation factors used to derive national BAFs can be measured or predicted using some or all of the following four methods, depending on the type of chemical and its properties. These methods are:

(1) a measured BAF obtained from a field study (i.e., a field-measured BAF);

(2) a BAF predicted from a field-measured BSAF;

(3) a BAF predicted from a laboratory-measured BCF (with or without adjustment by an FCM); and

(4) a BAF predicted from a chemical's octanol-water partition coefficient (K_{ow}), with or without adjustment using an FCM.

A brief summary of each of the four methods is provided below. Additional details on the use of these four methods is provided in Section 5.4 (for nonionic organics), Section 5.5 (for ionic organics) and Section 5.6 (for inorganics and organometallics).

1. **Field-Measured BAF.** Use of a field-measured BAF, which is the most direct measure of bioaccumulation, is the only method that can be used to derive a national BAF for all types of chemicals (i.e., nonionic organic, ionic organic, and inorganic and organometallic chemicals). A field-measured BAF is determined from a field study using measured chemical concentrations in the aquatic organism and its surrounding water. Because field studies are conducted in natural aquatic ecosystems, a field-measured BAF reflects an organism's exposure to a chemical through all relevant exposure pathways (i.e., water, sediment, and diet). A field-measured BAF also reflects any metabolism of a chemical that might occur in the aquatic organism or its food web. Therefore, field-measured BAFs are appropriate for all chemicals, regardless of the extent of chemical metabolism in biota.

2. **Field-measured BSAF.** For nonionic organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies), a BAF can also be predicted from BSAFs. A BSAF is similar to a field-measured BAF in that the concentration of a chemical in biota is measured in the field and reflects an organism's exposure to all relevant exposure routes. A BSAF also reflects any chemical metabolism that might occur in the aquatic organism or its food web. However, unlike a field-measured BAF which references the biota concentration to the water concentration, a BSAF references the biota concentration to the sediment concentration. Use of the BSAF procedure is restricted to organic chemicals which are classified as being moderately to highly hydrophobic.

3. **Lab-measured BCF.** A laboratory-measured BCF can also be used to estimate a BAF for organic and inorganic chemicals. However, unlike a field-measured BAF or a BAF predicted from a field-measured BSAF, a laboratory-measured BCF only reflects the accumulation of chemical through the water exposure route. Laboratory-measured BCFs may therefore under estimate BAFs for chemicals where accumulation from sediment or dietary sources is important. In these cases, laboratory-measured BCFs can be multiplied by a FCM to reflect accumulation from non-aqueous (i.e., food chain) pathways of exposure. Since a laboratory-measured BCF is determined using the measured concentration of a chemical in an aquatic organism and its surrounding water, a laboratory-measured BCF reflects any metabolism of the chemical that occurs in the organism, but not in the food web.

4. **K_{ow}.** A chemical's octanol-water partition coefficient, or K_{ow}, can also be used to predict a BAF for nonionic organic chemicals. This procedure is appropriate only for nonionic

organic chemicals (and certain ionic organic chemicals where similar lipid and organic carbon partitioning behavior applies). The K_{ow} has been extensively correlated with the BCF for nonionic organic chemicals that are poorly metabolized by aquatic organisms. Therefore, where substantial metabolism is known to occur in biota, the K_{ow} is not used to predict the BAF. For nonionic organic chemicals where chemical exposure through the food web is important, use of the K_{ow} alone will under predict the BAF. In such cases, the K_{ow} is adjusted with a FCM similar to the BCF procedure above.

5.3.2 Overview of BAF Derivation Framework

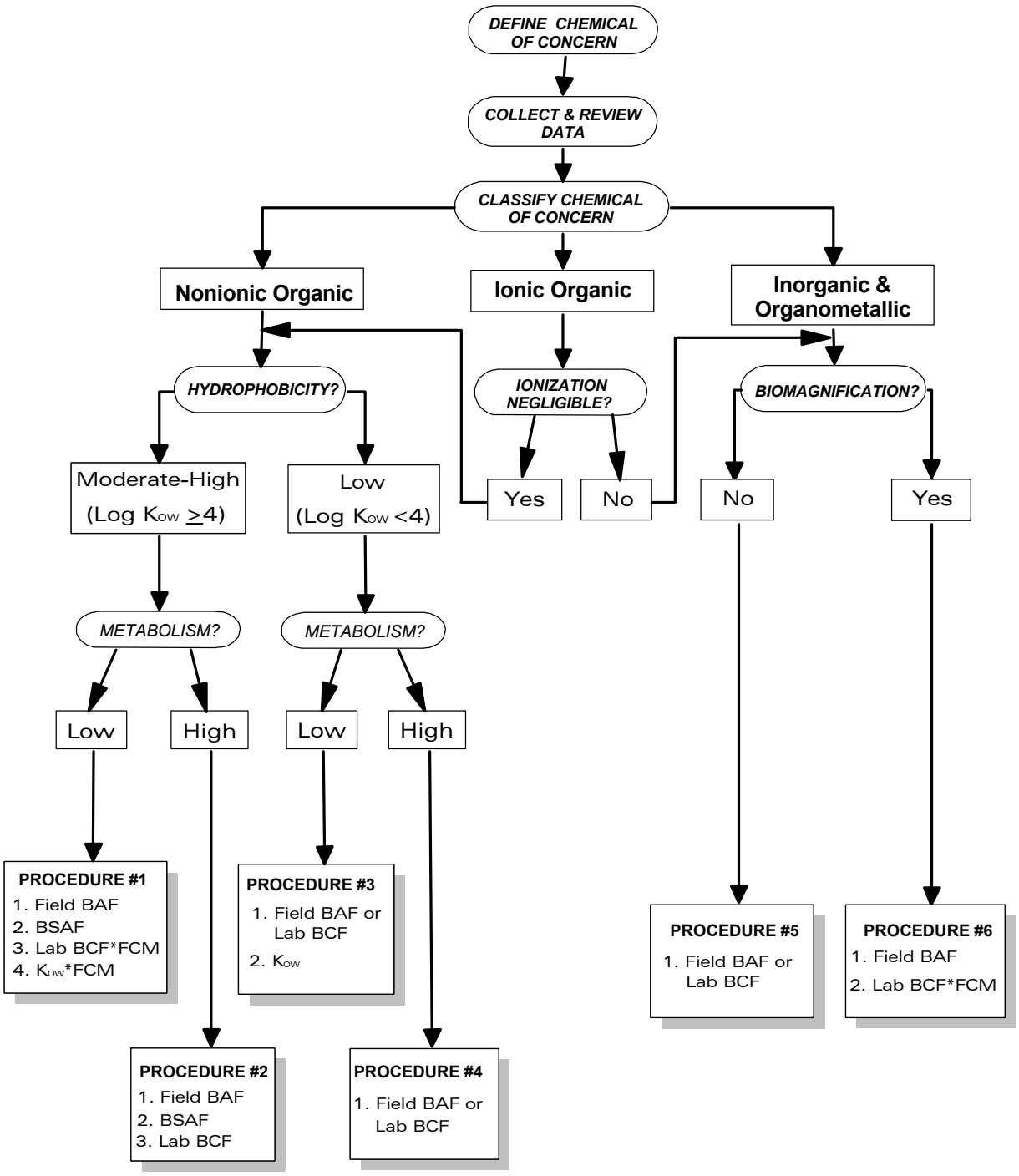
Although up to four methods can be used to derive a BAF as described in the previous section, it is evident that these methods do not apply equally to all types of chemicals. In addition, experience demonstrates that the required data will usually not be available to derive a BAF value using all of the applicable methods. As a result, EPA has developed the following guidelines to direct users in selecting the most appropriate method(s) for deriving a national BAF.

Figure 5-1 shows the overall framework of EPA's national BAF methodology. This framework illustrates the major steps and decisions that will ultimately lead to calculating a national BAF using one of six hierarchical procedures shown at the bottom of Figure 5-1. Each procedure contains a hierarchy of the BAF derivation methods discussed above, the composition of which depends on the chemical type and certain chemical properties (e.g., its degree of hydrophobicity and expected degree of metabolism and biomagnification). The number assigned to each BAF method within a procedure indicates its general order of preference for deriving a national BAF value. The goal of the framework and accompanying guidelines is to enable full use of available data and methods for deriving a national BAF value while appropriately restricting the use of certain methods to reflect their inherent limitations.

The first step in the framework is to define the chemical of concern. As described in Section 5.3.3, the chemical used to derive the national BAF should be consistent with the chemical used to derive the critical health assessment value. The second step is to collect and review all relevant data on bioconcentration and bioaccumulation of the chemical of concern (see Section 5.3.4). Once pertinent data are reviewed, the third step is to classify the chemical of concern into one of three broadly defined chemical categories: (1) nonionic organic chemicals, (2) ionic organic chemicals, and (3) and inorganic and organometallic chemicals. Guidance for classifying chemicals into these three categories is provided in Section 5.3.5.

After a chemical has been classified into one of the three categories, other information is used to select one of six hierarchical procedures to derive the national BAF. The specific procedures for deriving a BAF for each chemical group are discussed in Section 5.4 for nonionic organics, Section 5.5 for ionic organics, and Section 5.6 for inorganics and organometallics.

Figure 5-1. Framework for Deriving a National BAF



Detailed guidance concerning the first three steps of the derivation process (i.e, defining the chemical of concern, collecting and reviewing data, and classifying the chemical of concern) is provided in the following three sections.

5.3.3 Defining the Chemical of Concern

Defining the chemical of concern is the first step in deriving a national BAF. This step involves precisely defining the form(s) of the chemical upon which the national BAF value will be derived. Although this step is usually straightforward for single chemicals, complications can arise when the chemical of concern occurs as a mixture. The following guidelines should be followed for defining the chemical of concern.

1. Information for defining the chemical of concern should be obtained from the health and exposure assessment portions of the criteria derivation effort. The chemical(s) used to derive the national BAF should be consistent with the chemical(s) used to derive the reference dose (RfD), point of departure/uncertainty factor (POD/UF), or cancer potency factor.

2. In most cases, the RfD, POD/UF, or cancer potency factor will be based on a single chemical. In some cases, the RfD, POD/UF, or cancer potency factor will be based on a mixture of compounds, typically within the same chemical class (e.g., toxaphene, chlordane). In these situations, the national BAF should be derived in a manner that is consistent with the mixture used to express the health assessment.

    a. If sufficient data are available to reliably assess the bioaccumulation of each relevant compound contained in the mixture, then the national BAF(s) should be derived using the BAFs for the individual compounds of the mixture and appropriately weighted to reflect the mixture composition used to establish the RfD, POD/UF, or cancer potency factor. An example of this approach is shown in the derivation of BAFs for PCBs in the GLI Rulemaking (USEPA, 1997).

    b. If sufficient data are not available to reliably assess the bioaccumulation of individual compounds of the mixture, then the national BAF(s) should be derived using BAFs for the same or appropriately similar chemical mixture as that used to establish the RfD, POD/UF, or cancer potency value.

5.3.4 Collecting and Reviewing Data

The second step in deriving a national BAF is to collect and review all relevant bioaccumulation data for the chemical of concern. The following guidance should be followed for collecting and reviewing bioaccumulation data for deriving national BAFs.

1. All data on the occurrence and accumulation of the chemical of concern in aquatic animals and plants should be collected and reviewed for adequacy.

2. A comprehensive literature search strategy should be used for gathering bioaccumulation-related data. An example of a comprehensive literature search strategy is provided in the Bioaccumulation TSD.

3. All data that are used should contain sufficient supporting information to indicate that acceptable measurement procedures were used and that the results are probably reliable. In some cases it may be appropriate to obtain additional written information from the investigator.

4. Questionable data, whether published or unpublished, should not be used. Guidance for assessing the acceptability of bioaccumulation and bioconcentration studies is found in Sections 5.4, 5.5, and 5.6.

5.3.5 Classifying the Chemical of Concern

The next step in deriving a national BAF consists of classifying the chemical of concern into one of three categories: nonionic organic, ionic organic, and inorganic and organometallic (Figure 5-1). This step helps to determine which of the four methods described in Section 5.3.1 are appropriate for deriving BAFs. The following guidance applies for classifying the chemical of concern.

1. **Nonionic Organic Chemicals.** For the purposes of the 2000 Human Health Methodology, nonionic organic chemicals are those organic compounds that do not ionize substantially in natural bodies of water. These chemicals are also referred to as neutral or nonpolar organics in the scientific literature. Due to their neutrality, nonionic organic chemicals tend to associate with other neutral (or near neutral) compartments in aquatic ecosystems (e.g., lipid, organic carbon). Examples of nonionic organic chemicals which have been widely studied in terms of their bioaccumulation include polychlorinated biphenyls (PCBs), polychlorinated dibenzo-p-dioxins and furans, many chlorinated pesticides, and polynuclear aromatic hydrocarbons (PAHs). Procedures for deriving a national BAF for nonionic organic chemicals are provided in Section 5.4.

2. **Ionic Organic Chemicals.** For the purposes of the 2000 Human Health Methodology, ionic organic chemicals are considered to include those chemicals that contain functional groups with exchangeable protons such as hydroxyl, carboxylic, and sulfonic groups and functional groups that readily accept protons such as amino and aromatic heterocyclic nitrogen (pyridine) groups. Ionic organic chemicals undergo ionization in water, the extent of which depends on pH and the pKa of the chemical. Because the ionized species of these chemicals behave differently from the neutral species, separate guidance is provided for deriving BAFs for ionic organic chemicals. Procedures for deriving national BAFs for ionic organic chemicals are provided in Section 5.5.

3. **Inorganic and Organometallic Chemicals.** The inorganic and organometallic category is considered to include inorganic minerals, other inorganic compounds and elements,

metals (e.g., copper, cadmium, chromium, zinc), metalloids (selenium, arsenic) and organometallic compounds (e.g., methylmercury, tributyltin, tetraalkyllead). Procedures for deriving BAFs for inorganic and organometallic chemicals are provided in Section 5.6.

5.4 NATIONAL BIOACCUMULATION FACTORS FOR NONIONIC ORGANIC CHEMICALS

5.4.1 Overview

This section contains the methodology for deriving national BAFs for nonionic organic chemicals as defined in Section 5.3.5. The four general steps of this methodology are:

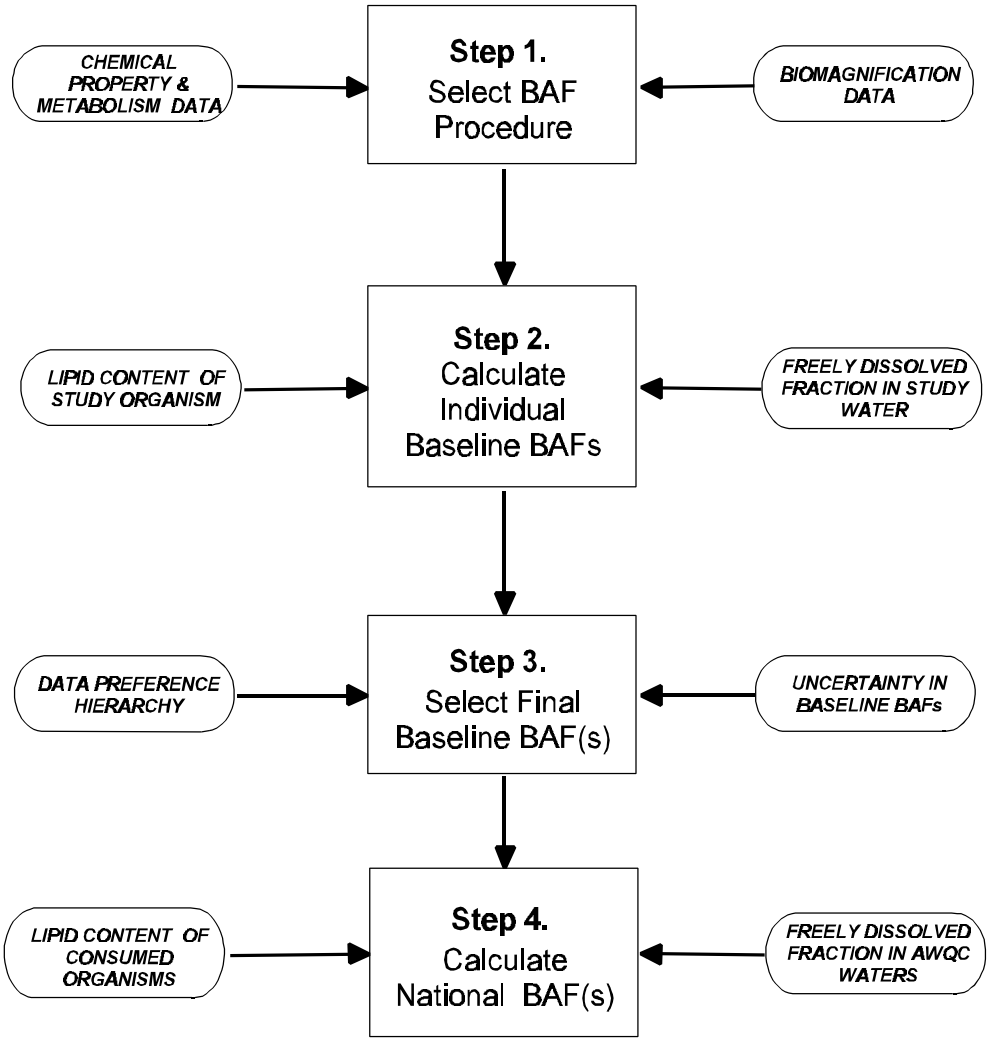
1. Selecting the BAF derivation procedure,
2. Calculating individual baseline BAF_R^{fd}s,
3. Selecting the final baseline BAF_R^{fd}s, and
4. Calculating the national BAFs from the final baseline BAF_R^{fd}s.

A schematic of this four-step process is shown in Figure 5-2.

Step 1 of the methodology (selecting the BAF derivation procedure) determines which of the four BAF procedures summarized in Figure 5-1 will be appropriate for deriving the national BAF. Step 2 involves calculating individual, species-specific BAF_R^{fd}s using all of the methods available within the selected BAF derivation procedure. Calculating the individual baseline BAF_R^{fd}s involves using data from the field site or laboratory where the original data were collected to account for site-specific factors which affect the bioavailability of the chemical to aquatic organisms (e.g., lipid content of study organisms and freely dissolved concentration in study water). Step 3 of the methodology consists of selecting the final baseline BAF_R^{fd}s from the individual baseline BAF_R^{fd}s by taking into account the uncertainty in the individual BAFs and the data preference hierarchy selected in Step 1. The final step is to calculate a BAF (or BAFs) that will be used in the derivation of 304(a) criteria (i.e., referred to as the national BAF). This step involves adjusting the final baseline BAF_R^{fd}(s) to reflect certain factors that affect bioavailablity of the chemical to aquatic organisms in waters to which the national 304(a) criteria will apply (e.g., the freely dissolved fraction expected in U.S. waters and the lipid content of consumed aquatic organisms). Baseline BAF_R^{fd}s are not used directly in the derivation of the 304(a) criteria because they do not reflect the conditions that affect bioavailability in U.S. waters.

Section 5.4.2 below provides detailed guidance for selecting the appropriate BAF derivation procedure (Step 1 of the process). Guidance on calculating individual baseline BAF_R^{fd}s, selecting the final baseline BAF, and calculating the national BAF (Steps 2 through 4 of the process) is provided in separate sections under each of the four BAF derivation procedures.

Figure 5-2. BAF Derivation for Nonionic Organic Chemicals



5.4.2 Selecting the BAF Derivation Procedure

This section describes the decisions that should be made to select one of the four available hierarchical procedures for deriving a national BAF for nonionic organic chemicals (Procedures #1 through #4 of Figure 5-1). As shown in Figure 5-1, two decision points exist in selecting the BAF derivation procedure. The first decision point requires knowledge of the chemical's hydrophobicity (i.e., the K_{ow} of the chemical). Guidance for selecting the K_{ow} for a chemical is provided in the Bioaccumulation TSD. The K_{ow} provides an initial basis for assessing whether biomagnification may be a concern for nonionic organic chemicals. The second decision point is based on the rate of metabolism for the chemical in the target organism. Guidance for assessing whether a high or low rate of metabolism is likely for a chemical of concern is provided below in Section 5.4.2.3. With the appropriate information for these two decision points, the BAF derivation procedure should be selected using the following guidelines.

5.4.2.1 Chemicals with Moderate to High Hydrophobicity

1. For the purposes of the 2000 Human Health Methodology, nonionic organic chemicals with log K_{ow} values equal to or greater than 4.0 should be classified as moderately to highly hydrophobic. For moderately to highly hydrophobic nonionic organic chemicals, available data indicate that exposure through the diet and other non-aqueous routes can become important in determining chemical residues in aquatic organisms (e.g., Russell et al., 1999; Fisk et al., 1998; Oliver and Niimi, 1983; Oliver and Niimi, 1988; Niimi, 1985; Swackhammer and Hites, 1988). Dietary and other non-aqueous exposure can become extremely important for those nonionic organic chemicals that are poorly metabolized by aquatic biota (e.g., certain PCB congeners, chlorinated pesticides, and polychlorinated dibenzo-p-dioxins and furans).

2. **Procedure #1** should be used to derive national BAFs for moderately to highly hydrophobic nonionic organic chemicals in cases where:

   (a) the rate of chemical metabolism by target aquatic organisms is expected to be sufficiently low such that biomagnification is of concern, or

   (b) the rate of chemical metabolism by target aquatic organisms is not sufficiently known.

   Procedure #1 accounts for non-aqueous exposure and the potential for biomagnification in aquatic food webs through the use of field-measured values for bioaccumulation (i.e., field measured BAF or BSAF) and FCMs when appropriate field data are unavailable. Guidance on deriving national BAFs using Procedure #1 is found below in Section 5.4.3.

3. **Procedure #2** should be used to derive the national BAFs for moderately to highly hydrophobic nonionic organic chemicals in cases where:

(a) the rate of chemical metabolism by target aquatic organisms is expected to be sufficiently high such that biomagnification is not of concern.

Procedure #2 relaxes the requirement of using FCMs and eliminates the use of K_{ow}-based estimates of the BAF, two procedures that are most appropriate for poorly metabolized nonionic organic chemicals. Guidance on deriving national BAFs using Procedure #2 is found below in Section 5.4.4.

5.4.2.2 Chemicals with Low Hydrophobicity

1. For the purposes of these guidelines, nonionic organic chemicals with log K_{ow} values less than 4.0 should be classified as exhibiting low hydrophobicity. For nonionic organic chemicals that exhibit low hydrophobicity (i.e., log $K_{ow} < 4.0$), available information indicates that non-aqueous exposure to these chemicals is not likely to be important in determining chemical residues in aquatic organisms (e.g., Fisk et al., 1998; Gobas et al., 1993; Connolly and Pedersen, 1988; Thomann, 1989). For this group of chemicals, laboratory-measured BCFs and K_{ow}-predicted BCFs do not require adjustment with FCMs for determining the national BAF (Procedures #3 and #4), unless other appropriate data indicate differently.

   Other appropriate data include studies clearly indicating that non-aqueous exposure is important such that use of a BCF would substantially underestimate residues in aquatic organisms. In these cases, Procedure #1 should be used to derive the BAF for nonionic organic chemicals with log $K_{ow} < 4.0$. Furthermore, the data supporting the K_{ow} determination should be carefully reviewed for accuracy and appropriate interpretation, since the apparent discrepancy may be due to errors in determining K_{ow}.

2. **Procedure #3** should be used to derive national BAFs for nonionic organic chemicals of low hydrophobicity in cases where:

   (a) the rate of chemical metabolism by target aquatic organisms is expected to be negligible, such that tissue residues of the chemical of concern are not substantially reduced compared to an assumption of no metabolism, or

   (b) the rate of chemical metabolism by target aquatic organisms is not sufficiently known.

   Procedure #3 includes the use of K_{ow}-based estimates of the BCF to be used when lab or field data are absent. Guidance on deriving national BAFs using Procedure #3 is found below in Section 5.4.5.

3. **Procedure #4** should be used to derive national BAFs for nonionic organic chemicals of low hydrophobicity in cases where:

(a) the rate of chemical metabolism by target aquatic organisms is expected to be sufficiently high, such that tissue residues of the chemical of concern are substantially reduced compared with an assumption of no metabolism.

Procedure #4 eliminates the option of using K_{ow}-based estimates of the BAF because the K_{ow} may over-predict accumulation when a chemical is metabolized substantially by an aquatic organism. Guidance on deriving national BAFs using Procedure #4 is found below in Section 5.4.6.

5.4.2.3 Assessing Metabolism

Currently, assessing the degree to which a chemical is metabolized by aquatic organisms is confounded by a variety of factors. First, conclusive data on chemical metabolism in aquatic biota are largely lacking. Such data include whole organism studies where the metabolic rates and breakdown products are quantified in fish and other aquatic organisms relevant to human consumption. However, the majority of information on metabolism is derived from *in vitro* liver microsomal preparations in which primary and secondary metabolites may be identified and their rates of formation may or may not be quantified. Extrapolating results from *in vitro* studies to the whole organism involves considerable uncertainty. Second, there are no generally accepted procedures for reliably predicting chemical metabolism by aquatic organisms in the absence of measured data. Third, the rate at which a chemical is metabolized by aquatic organisms can be species and temperature dependent. For example, PAHs are known to be metabolized readily by vertebrate aquatic species (primarily fish), although at rates much less than those observed for mammals. However, the degree of metabolism in invertebrate species is generally much less than the degree in vertebrate species (James, 1989). One hypothesis for this difference is that the invertebrate species lack the detoxifying enzymes and pathways that are present in many vertebrate species.

Given the current limitations on assessing the degree of chemical metabolism by aquatic organisms, the assessment of metabolism should be made on a case-by-case basis using a weight-of-evidence approach. When assessing a chemical's likelihood to undergo substantial metabolism in a target aquatic organism, the following data should be carefully evaluated:

(1) *in vivo* chemical metabolism data,
(2) bioconcentration and bioaccumulation data,
(3) data on chemical occurrence in target aquatic biota, and
(4) *in vitro* chemical metabolism data.

1. ***In vivo* Data.** *In vivo* data on metabolism in aquatic organisms are from studies of chemical metabolism using whole organisms. These studies are usually conducted using large fish from which blood, bile, urine, and individual tissues can be collected for the identification and quantification of metabolites formed over time. *In vivo* studies are considered the most useful for evaluating a chemical's degree of metabolism in an organism because both oxidative (Phase I) and conjugative (Phase II) metabolism can be

assessed in these studies. Mass-balance studies, in which parent compound elimination is quantified separately from biotransformation and elimination of metabolites, allow calculation of conversion rate of parent to metabolite as well as metabolite elimination. This information might be used to estimate loss due to metabolism separately from that due to elimination of the parent compound for adjustment of K_{ow}-predicted BAFs. However, due to the analytical and experimental challenges these studies pose, data of this type are limited. Less rigorous *in vivo* metabolism studies might include the use of metabolic blockers to demonstrate the influence of metabolism on parent compound kinetics. However, caution should be used in interpretation of absolute rates from these data due to the lack of specificity of mammalian derived blockers in aquatic species (Miranda et al., 1998).

2. **Bioconcentration or Bioaccumulation Data.** Data on chemical bioconcentration or bioaccumulation in aquatic organisms can be used indirectly for assessing metabolism. This assessment involves comparing acceptable lab-measured BCFs or field-measured BAFs (after converting to baseline values using procedures below) with the chemical's predicted value based on K_{ow}. The theoretical basis of bioconcentration and bioaccumulation for nonionic organic chemicals indicates that a chemical's baseline BCF should be similar to its K_{ow}-predicted value if metabolism is not occurring or is minimal (see the Bioaccumulation TSD). This theory also indicates that baseline BAFs should be similar to or higher than the K_{ow} for poorly metabolized organic chemicals, with highly hydrophobic chemicals often exhibiting higher baseline BAFs than K_{ow} values. Thus, if a chemical's baseline BCF or BAF is substantially lower than its K_{ow}, this may be an indication that the chemical is being metabolized by the aquatic organism of concern. Note, however, that this difference may also indicate problems in the experimental design or analytical chemistry, and that it may be difficult to discern the difference.

3. **Chemical Occurrence Data.** Although by no means definitive, data on the occurrence of chemicals in aquatic biota (i.e., residue studies) may offer another useful line of evidence for evaluating a chemical's likelihood to undergo substantial metabolism. Such studies are most useful if they have been conducted repeatedly over time and over wide geographical areas. Such studies might indicate a chemical is poorly metabolized if data show that the chemical is being biomagnified in the aquatic food web (i.e., higher lipid-normalized residues in successive trophic levels). Conversely, such studies might indicate a chemical is being metabolized substantially if residue data show a decline in residues with increasing trophic level. Again, other reasons for increases or decreases in concentrations with increasing trophic level might exist and should be carefully evaluated (e.g., incorrect food web assumptions, differences in exposure concentrations).

4. ***In vitro* Data.** *In vitro* metabolism data include data from studies where specific sub-cellular fractions (e.g., microsomal, cytosolic), cells, or tissues from an organism are tested outside the body (i.e., in test-tubes, cell- or tissue-culture). Compared with *in vivo* studies of chemical metabolism in aquatic organisms, *in vitro* studies are much more plentiful in the literature, with the majority of studies characterizing oxidative (Phase I)

reactions de-coupled from conjugative (Phase II) metabolism. Cell, tissue, or organ level *in vitro* studies are less common but provide a more complete assessment of metabolism. While such studies are particularly useful for identifying the pathways, rates of formation, and metabolites formed, as well as the enzymes involved and differences in the temperature dependence of metabolism across aquatic species, they suffer from uncertainty when results are extrapolated to the whole organism. This uncertainty results from the fact that dosimetry (i.e., delivery of the toxicant to, and removal of metabolite from, the target tissue) cannot currently be adequately reproduced in the laboratory or easily modeled.

When assessing chemical metabolism using the above information, the following guidelines apply.

a. A finding of substantial metabolism should be supported by two or more lines of evidence identified using the data described above.

b. At least one of the lines of evidence should be supported by either *in vivo* metabolism data or acceptable bioconcentration or bioaccumulation data.

c. A finding of substantial metabolism in one organism should not be extrapolated to another organism or another group of organisms unless data indicate similar metabolic pathways exist (or are very likely to exist) in both organisms. *In vitro* data may be particularly useful in cross-species extrapolations.

d. Finally, in situations where sufficient data are not available to properly assess the likelihood of significant metabolism in aquatic biota of concern, the chemical should be assumed to undergo little or no metabolism. This assumptions reflects a policy decision by EPA to err on the side of public health protection when sufficient information on metabolism is lacking.

5.4.3 Deriving National BAFs Using Procedure #1

This section contains guidance for calculating national BAFs for nonionic organic chemicals using Procedure #1 shown in Figure 5-1. The types of nonionic organic chemicals for which Procedure #1 is most appropriate are those that are classified as moderately to highly hydrophobic and subject to low (or unknown) rates of metabolism by aquatic biota (see Section 5.4.2 above). Non-aqueous contaminant exposure and subsequent biomagnification in aquatic food webs are of concern for chemicals that are classified in this category. Some examples of nonionic organic chemicals for which Procedure #1 is considered appropriate include:

- tetra-, penta- & hexachlorobenzenes;
- PCBs;
- octachlorostyrene;
- hexachlorobutadiene;

- endrin, dieldrin, aldrin;
- mirex, photomirex;
- DDT, DDE, DDD; and
- heptachlor, chlordane, nonachlor.

Under Procedure #1, the following four methods may be used in deriving a national BAF:

- using a BAF from an acceptable field study (i.e., a field-measured BAF);
- predicting a BAF from an acceptable field-measured BSAF;
- predicting a BAF from an acceptable laboratory-measured BCF and FCM; and
- predicting a BAF from an acceptable K_{ow} and FCM.

As shown in Figure 5-2, once the derivation procedure has been selected, the next steps in deriving a national BAF for a given trophic level include: calculating individual baseline BAF_R^{fd}s (step 2), selecting the final baseline BAF_R^{fd} (step 3), and calculating the national BAF from the final baseline BAF_R^{fd} (step 4). Each of these three steps is discussed separately below.

5.4.3.1 Calculating Individual Baseline BAF_R^{fd}s

Calculating an individual baseline BAF_R^{fd} involves normalizing the field-measured BAF_T^t (or laboratory-measured BCF_T^t) which are based on total concentrations in tissue and water by the lipid content of the study organisms and the freely dissolved concentration in the study water. Both the lipid content in the organism and the freely dissolved concentration (as influenced by organic carbon in water) have been shown to be important factors that influence the bioaccumulation of nonionic organic chemicals (e.g., Mackay, 1982; Connolly and Pederson, 1988; Thomann, 1989, Suffet et al., 1994). Therefore, baseline BAF_R^{fd}s (which are expressed on a freely dissolved and lipid-normalized basis) are considered more amenable to extrapolating between different species and bodies of water compared to BAFs expressed using the total concentration in the tissue and water. Because bioaccumulation can be strongly influenced by the trophic position of aquatic organisms (either due to biomagnification or physiological differences), extrapolation of baseline BAF_R^{fd}s should not be performed between species of different trophic levels.

1. For each species for which acceptable data are available, calculate all possible baseline BAF_R^{fd}s using each of the four methods shown above for Procedure #1.

2. Individual baseline BAF_R^{fd}s should be calculated from field-measured BAF_T^ts, field-measured BSAFs, laboratory BCF_T^ts, and the K_{ow} according to the following procedures.

A. Baseline BAF_R^{fd}s from Field-Measured BAFs

A baseline BAF_R^{fd} should be calculated from each field-measured BAF_T^t using information on the lipid fraction in the tissue of concern for the study organism and the fraction of the total chemical that is freely dissolved in the study water.

1. **Baseline BAF_R^{fd} Equation.** For each acceptable field-measured BAF_T^t, calculate a baseline BAF_R^{fd} using the following equation:

$$\text{Baseline } BAF_\ell^{fd} = \left[\frac{\text{Measured } BAF_T^t}{f_{fd}} - 1 \right] \left(\frac{1}{f_\ell} \right) \qquad \text{(Equation 5-10)}$$

where:

Baseline BAF_R^{fd}	=	BAF expressed on a freely dissolved and lipid-normalized basis
Measured BAF_T^t	=	BAF based on total concentration in tissue and water
f_R	=	Fraction of the tissue that is lipid
f_{fd}	=	Fraction of the total chemical that is freely dissolved in the ambient water

The technical basis of Equation 5-10 is provided in the Bioaccumulation TSD. Guidance for determining each component of Equation 5-10 is provided below.

2. **Determining the Measured BAF_T^t.** The field-measured BAF_T^t shown in Equation 5-10 should be calculated based on the total concentration of the chemical in the appropriate tissue of the aquatic organism and the total concentration of the chemical in ambient water at the site of sampling. The equation to derive a measured BAF_T^t is:

$$\text{Measured } BAF_T^t = \frac{C_t}{C_w} \qquad \text{(Equation 5-11)}$$

where:

C_t	=	Total concentration of the chemical in the specified wet tissue
C_w	=	Total concentration of chemical in water

The data used to calculate a field-measured BAF_T^t should be reviewed thoroughly to assess the quality of the data and the overall uncertainty in the BAF value. The following general criteria apply in determining the acceptability of field-measured BAFs that are being considered for deriving national BAFs using Procedure #1.

a. Aquatic organisms used to calculate a field-measured BAF_T^t should be representative of aquatic organisms that are commonly consumed in the United States. An aquatic organism that is not commonly consumed in the United States can be used to calculate an acceptable field-measured BAF_T^t provided that the

organism is considered to be a reasonable surrogate for a commonly consumed organism. Information on the ecology, physiology, and biology of the organism should be reviewed when assessing whether an organism is a reasonable surrogate of a commonly consumed organism.

b. The trophic level of the study organism should be determined by taking into account its life stage, diet, size, and the food web structure at the study location. Information from the study site (or similar sites) is preferred when evaluating trophic status. If such information is lacking, general information for assessing trophic status of aquatic organisms can be found in USEPA (2000a,b,c).

c. The percent lipid of the tissue used to determine the field-measured BAF_T^t should be either measured or reliably estimated to permit lipid-normalization of the chemical's tissue concentration.

d. The study from which the field-measured BAF_T^t is derived should contain sufficient supporting information from which to determine that tissue and water samples were collected and analyzed using appropriate, sensitive, accurate, and precise analytical methods.

e. The site of the field study should not be so unique that the BAF cannot be reasonably extrapolated to other locations where the BAF and resulting criteria will apply.

f. The water concentration(s) used to derive the BAF should reflect the average exposure of the aquatic organism that corresponds to the concentration measured in its tissue of concern. For nonionic organic chemicals, greater temporal and spatial averaging of chemical concentrations is required as the K_{ow} increases. In addition, as variability in water concentrations increase, greater temporal and spatial averaging is also generally required. Greater spatial averaging is also generally required for more mobile organisms.

g. The concentrations of particulate organic carbon and dissolved organic carbon in the study water should be measured or reliably estimated.

EPA is currently developing guidance for designing and conducting field studies for determining field-measured BAF_T^ts, including recommendations for minimum data requirements. A more detailed discussion of factors that should be considered when determining field-measured BAF_T^ts is provided in the Bioaccumulation TSD.

3. **Determining the Fraction Freely Dissolved (f_{fd}).** As illustrated by Equation 5-10, the fraction of the nonionic organic chemical that is freely dissolved in the study water is required for calculating a baseline BAF_R^{fd} from a field-measured BAF_T^t. The freely dissolved fraction is the portion of the nonionic organic chemical that is not bound to

particulate organic carbon or dissolved organic carbon. Together, the concentration of a nonionic organic chemical that is freely dissolved, bound to dissolved organic carbon, and bound to particulate organic carbon constitute its total concentration in water. As discussed further in the Bioaccumulation TSD, the freely dissolved fraction of a chemical is considered to be the best expression of the bioavailable form of nonionic organic chemicals to aquatic organisms (e.g., Suffet et al., 1994; USEPA, 1995b). Because the fraction of a nonionic organic chemical that is freely dissolved may vary among different bodies of water as a result of differences in dissolved and particulate organic carbon in the water, the bioavailability of the total chemical concentration in water is expected to vary from one body of water to another. Therefore, BAFs which are based on the freely dissolved concentration in water (rather than the total concentration in water) are considered to be more reliable for extrapolating and aggregating BAFs among different bodies of water. Currently, availability of BAFs based on measured freely dissolved concentrations is very limited, partly because of difficulties in analytically measuring the freely dissolved concentration. Thus, if a BAF based on the total water concentration is reported in a given study, the fraction of the chemical that is freely dissolved should be predicted using information on the organic carbon content in the study water.

a. **Equation for Determining the Freely Dissolved Fraction.** If reliable measured data are unavailable to directly determine the freely dissolved fraction of the chemical in water, the freely dissolved fraction should be estimated using the following equation.

$$f_{fd} = \frac{1}{[1 + (POC \cdot K_{ow}) + (DOC \cdot 0.08 \cdot K_{ow})]} \qquad \text{(Equation 5-12)}$$

where:

POC	=	concentration of particulate organic carbon (kg/L)
DOC	=	concentration of dissolved organic carbon (kg/L)
K_{ow}	=	n-octanol water partition coefficient for the chemical

In Equation 5-12, K_{ow} is being used to estimate the partition coefficient to POC (i.e., K_{POC} in L/kg) and $0.08 \cdot K_{ow}$ is being used to estimate the partition coefficient to DOC (i.e., the K_{DOC} in L/kg). A discussion of the technical basis, assumptions, and uncertainty associated with the derivation and application of Equation 5-12 is provided in the Bioaccumulation TSD.

b. **POC and DOC Values.** When converting from the total concentration of a chemical to a freely dissolved concentration using Equation 5-12 above, the POC and DOC concentrations should be obtained from the original study from which the field-measured BAF is determined. If POC and DOC concentrations are not reported in the BAF study, reliable estimates of POC and DOC might be obtained

from other studies of the same site used in the BAF study or closely related site(s) within the same water body. When using POC/DOC data from other studies of the same water body, care should be taken to ensure that environmental and hydrological conditions that might affect POC or DOC concentrations (i.e., runoff events, proximity to ground water or surface water inputs, sampling season) are reasonably similar to those in the BAF study. Additional information related to selecting POC and DOC values is provided in the Bioaccumulation TSD.

In some cases, BAFs are reported using the concentration of the chemical in filtered or centrifuged water. When converting these BAFs to a freely dissolved basis, the concentration of POC should be set equal to zero when using Equation 5-12. Particulates are removed from water samples by filtering or centrifuging the sample.

c. **Selecting K_{ow} Values.** A variety of techniques are available to measure or predict K_{ow} values. The reliability of these techniques depends to a large extent on the K_{ow} of the chemical. Because K_{ow} is an important input parameter for calculating the freely dissolved concentration of nonionic organic chemicals and for deriving BAFs using the other three methods of Procedure #1, care should be taken in selecting the most reliable K_{ow} value. The value of K_{ow} for use in estimating the freely dissolved fraction and other procedures used to derive national BAFs should be selected based on the guidance presented in the Bioaccumulation TSD.

4. **Determining the Fraction Lipid (f_R).** Calculating a baseline BAF_R^{fd} for a nonionic organic chemical using Equation 5-10 also requires that the total chemical concentration measured in the tissue used to determine the field-measured BAF_T^t be normalized by the lipid fraction (f_R) in that same tissue. Lipid normalization of tissue concentrations reflects the assumption that BAFs (and BCFs) for nonionic organic chemicals are directly proportional to the percent lipid in the tissue upon which they are based. This assumption means that an organism with a two percent lipid content would be expected to accumulate twice the amount of a chemical at steady state compared with an organism with one percent lipid content, all else being equal. The assumption that aquatic organisms accumulate nonionic organic chemicals in proportion to their lipid content has been extensively evaluated in the literature (Mackay, 1982; Connell, 1988; Barron, 1990) and is generally accepted. Because the lipid content in aquatic organisms can vary both within and across species, BAFs that are expressed using the lipid-normalized concentration (rather than the total concentration in tissue) are considered to be the most reliable for aggregating multiple BAF values for a given species. Additional discussion of technical basis, assumptions, and uncertainties involved in lipid normalization is provided in the Bioaccumulation TSD.

a. The lipid fraction f_R, is routinely reported in bioaccumulation studies involving nonionic organic chemicals. If the lipid fraction is not reported in the BAF study,

it can be calculated using the following equation if the appropriate data are reported:

$$f_l = \frac{M_l}{M_t} \qquad \text{(Equation 5-13)}$$

where:

M_R = Mass of lipid in specified tissue
M_t = Mass of specified tissue (wet weight)

b. Because lipid content can vary within an aquatic organism (and among tissues within that organism) due to several factors including the age and sex of the organism, changes in dietary composition, season of sampling and reproductive status, the lipid fraction used to calculate a baseline BAF_R^{fd} should be measured in the same tissue and organisms used to determine the field-measured BAF_T^t, unless comparability is demonstrated across organisms.

c. Experience has shown that different solvent systems used to extract lipids for analytical measurement can result in different quantities of lipids being extracted and measured in aquatic organisms (e.g., Randall et al.,1991, 1998). As a result, lipid measurements determined using different solvent systems might lead to apparent differences in lipid-normalized concentrations and lipid-normalized BAFs. The extent to which different solvent systems might affect lipid extractions (and lipid-normalized concentrations) is thought to vary depending on the solvent, chemical of concern, and lipid composition of the tissue being extracted. Guidance on measurement of lipid content, including the choice of solvent system and how different solvent systems may affect lipid content, is provided in the Bioaccumulation TSD.

B. Baseline BAF_R^{fd} Derived from BSAFs

The second method of determining a baseline BAF_R^{fd} for the chemical of concern in Procedure #1 involves the use of BSAFs. Although BSAFs may be used for measuring and predicting bioaccumulation directly from concentrations of chemicals in surface sediment, they may also be used to estimate BAFs (USEPA, 1995b; Cook and Burkhard, 1998). Since BSAFs are based on field data and incorporate effects of chemical bioavailability, food web structure, metabolism, biomagnification, growth, and other factors, BAFs estimated from BSAFs will incorporate the net effect of all these factors. The BSAF approach is particularly beneficial for developing water quality criteria for chemicals which are detectable in fish tissues and sediments, but are difficult to detect or measure precisely in the water column.

As shown by Equation 5-14 below, predicting baseline BAF_R^{fd}s using BSAFs requires that certain types of data be used for the chemicals of interest (for which BAFs are to be determined) and reference chemicals (for which BAFs are measured) from a common sediment-water-organism data set. Differences between BSAFs for different organic chemicals are good measures of the relative bioaccumulation potentials of the chemicals. When calculated from a common organism-sediment sample set, chemical-specific differences in BSAFs reflect the net effect of biomagnification, metabolism, food chain, bioenergetics, and bioavailability factors on the degree of each chemical's equilibrium/disequilibrium between sediment and biota. At equilibrium, BSAFs are expected to be approximately 1.0. However, deviations from 1.0 (reflecting disequilibrium) are common due to: conditions where water is not at equilibrium with surface sediment; differences in organic carbon content of water and sediment; kinetic limitations for chemical transfer between sediments and water associated with specific biota; biomagnification; or biological processes such as growth or biotransformation. BSAFs are most useful (i.e., most predictable from one site to another) when measured under steady-state (or near steady-state) conditions. The use of non-steady-state BSAFs, such as found with new chemical loadings or rapid increases in loadings, increases uncertainty in this method for the relative degree of disequilibrium between the reference chemicals and the chemicals of interest. In general, the fact that concentrations of hydrophobic chemicals in sediment are less sensitive than concentrations in water to fluctuations in chemical loading and distribution makes the BSAF method robust for estimating BAFs. Results from validation of the BAF procedure in Lake Ontario, the Fox River and Green Bay, Wisconsin, and the Hudson River, New York, demonstrate good agreement between observed and BSAF-predicted BAFs in the vast majority of comparisons made. Detailed results of the validation studies for the BSAF procedure are provided in the Bioaccumulation TSD.

Baseline BAF_R^{fd}s should be calculated using acceptable BSAFs for chemicals of interest and appropriate sediment-to-water fugacity (disequilibrium) ratios $(\Pi_{socw})_r/(K_{ow})_r$ for reference chemicals under the following guidelines.

1. **Baseline BAF_R^{fd} Equation.** For each species with an acceptable field measured $(BSAF)_I$, a baseline BAF_R^{fd} for the chemical of interest may be calculated using the following equation with an appropriate value of $(\Pi_{socw})_r/(K_{ow})_r$:

$$(Baseline\ BAF_l^{fd})_i = (BSAF)_i \frac{(D_{i/r})\ (\Pi_{socw})_r\ (K_{ow})_i}{(K_{ow})_r} \qquad \text{(Equation 5-14)}$$

where:

$(\text{Baseline } BAF_R^{fd})_I$	=	BAF expressed on a freely dissolved and lipid-normalized basis for chemical of interest "I"
$(BSAF)_I$	=	Biota-sediment accumulation factor for chemical of interest "I"

$(J_{socw})_r$ = sediment organic carbon to water freely dissolved concentration ratio of reference chemical "r"

$(K_{ow})_I$ = octanol-water partition coefficient for chemical of interest "I"

$(K_{ow})_r$ = octanol-water partition coefficient for the reference chemical "r"

$D_{i/r}$ = ratio between J_{socw} / K_{ow} for chemicals "I" and "r" (normally chosen so that $D_{i/r} = 1$)

The technical basis, assumptions, and uncertainties associated with Equation 5-14 are provided in the Bioaccumulation TSD. Guidance for determining each component of Equation 5-14 is provided below.

2. **Determining Field-Measured BSAFs.** BSAFs should be determined by relating lipid-normalized concentrations of chemicals in an organism (C_l) to organic carbon-normalized concentrations of the chemicals in surface sediment samples (C_{soc}) using the following equation:

$$BSAF = \frac{C_l}{C_{soc}} \qquad \text{(Equation 5-15)}$$

a. **Lipid-Normalized Concentration.** The lipid-normalized concentration of a chemical in an organism should be determined by:

$$C_l = \frac{C_t}{f_l} \qquad \text{(Equation 5-16)}$$

where:

C_t = Concentration of the chemical in the wet tissue (either whole organism or specified tissue) (µg/g)

f_R = Fraction lipid content in the tissue

b. **Organic Carbon-Normalized Concentration.** The organic carbon-normalized concentration of a chemical in sediment should be determined by:

$$C_{soc} = \frac{C_s}{f_{oc}} \qquad \text{(Equation 5-17)}$$

where:

C_s = Concentration of chemical in sediment (µg/g sediment)
f_{oc} = Fraction organic carbon in sediment

The organic carbon-normalized concentrations of the chemicals in surface sediment samples should be associated with the average exposure environment of the organism.

3. **Sediment-to-Water Partition Coefficient** (Π_{socw})$_r$. Sediment-to-water partition coefficients for reference chemicals should be determined by:

$$(\Pi_{socw})_r = \frac{(C_{soc})_r}{(C_w^{fd})_r} \qquad \text{(Equation 5-18)}$$

where:

$(C_{soc})_r$ = Concentration of a reference chemical in sediment normalized to sediment organic carbon
$(C_w^{fd})_r$ = Concentration of the reference chemical freely dissolved in water

4. **Selecting Reference Chemicals.** Reference chemicals with (Π_{socw}) / (K_{ow}) similar to that of the chemical of interest are preferred for this method. Theoretically, knowledge of the difference between sediment-to-water fugacity ratios for two chemicals, "I" and "r" ($D_{i/r}$), could be used when reliable reference chemicals that meet the fugacity equivalence condition are not available. Similarity of (Π_{socw}) / (K_{ow}) for two chemicals can be indicated on the basis of similar physical-chemical behavior in water (persistence, volatilization), similar mass loading histories, and similar concentration profiles in sediment cores.

Validation studies have demonstrated that choosing reference chemicals with well quantified concentrations in water is important because the uncertainty associated with measurement of barely detected chemicals is large (see the Bioaccumulation TSD). Similarity between K_{ow} values of the reference and target chemicals is generally desirable, although recent validation studies indicate that the accuracy of the method is not substantially decreased through use of reference chemicals with large differences in K_{ow}, as long as the chemicals are structurally similar and have similar persistence behavior in water and sediments.

5. The following data, procedural, and quality assurance requirements should be met for predicting baseline BAF_R^{fd}s using field-measured BSAFs:

a. Data on the reference chemicals and chemicals of interest should come from a common organism-water-sediment data set at a particular site.

b. The chemicals of interest and reference chemicals should have similar physicochemical properties and persistence in water and sediment.

c. The loadings history of the reference chemicals and chemicals of interest should be similar such that their expected sediment-water disequilibrium ratios (J_{socw}/K_{ow}) would not be expected to be substantially different (i.e., $D_{i/r} \sim 1$).

d. The use of multiple reference chemicals is generally preferred for determining the value of $(J_{socw})_r$ so long as the concentrations are well quantified and the aforementioned conditions for selecting reference chemicals are met. In some cases, use of a single reference chemical may be necessary because of limited data.

e. Samples of surface sediments (0-1 cm is ideal) should be from locations in which sediment is regularly deposited and is representative of average surface sediment in the vicinity of the organism.

f. The K_{ow} value for the target and reference chemicals should be selected as described in the Bioaccumulation TSD.

g. All other data quality and procedural guidelines described earlier for determining field-measured BAFs in Section 5.4.3.1(A) should be met.

Further details on the requirements for predicting BAFs from BSAF measurements, including the data, assumptions, and limitations of this approach are provided in the Bioaccumulation TSD.

C. Baseline BAF_R^{fd} from a Laboratory-Measured BCF_T^t and FCM

The third method in Procedure #1 consists of using a laboratory-measured BCF_T^t (i.e., a BCF based on total concentrations in tissue and water) and FCMs to predict a baseline BAF_R^{fd} for the chemical of concern. The BCF_T^t is used in conjunction with an FCM because non-aqueous routes of exposure and subsequent biomagnification is of concern for the types of chemicals applicable to Procedure #1. A laboratory-measured BCF inherently accounts for the effects of chemical metabolism that occurs in the organism used to calculate the BCF, but does not account for metabolism which may occur in other organisms of the aquatic food web.

1. **Baseline BAF_R^{fd} Equation.** For each acceptable laboratory-measured BCF_T^t, calculate a baseline BAF_R^{fd} using the following equation:

$$\text{Baseline } BAF_{\ell}^{fd} = (FCM) \cdot \left[\frac{\text{Measured } BCF_T^t}{f_{fd}} - 1 \right] \cdot \left(\frac{1}{f_{\ell}} \right) \qquad \text{(Equation 5-19)}$$

where:

Baseline BAF_R^{fd}	=	BAF expressed on a freely dissolved and lipid-normalized basis
Measured BCF_T^t	=	BCF based on total concentration in tissue and water
f_R	=	Fraction of the tissue that is lipid
f_{fd}	=	Fraction of the total chemical in the test water that is freely dissolved
FCM	=	The food chain multiplier either obtained from Table 5-1 by linear interpolation for the appropriate trophic level, or from appropriate field data

The technical basis for Equation 5-19 is provided in the Bioaccumulation TSD. Guidance for determining each component of Equation 5-19 is provided below.

2. **Determining the Measured BCF_T^t.** The laboratory-measured BCF_T^t shown in Equation 5-19 should be calculated using information on the total concentration of the chemical in the tissue of the organism and the total concentration of the chemical in the laboratory test water. The equation to derive a measured BCF_T^t is:

$$\text{Measured } BCF_T^t = \frac{C_t}{C_w} \qquad \text{(Equation 5-20)}$$

where:

C_t	=	Total concentration of the chemical in the specified wet tissue
C_w	=	Total concentration of chemical in the laboratory test water

The data used to calculate a laboratory-measured BCF_T^t should be reviewed thoroughly to assess the quality of the data and the overall uncertainty in the BCF value. The following general criteria apply in determining the acceptability of laboratory-measured BCF_T^t.

a. The test organism should not be diseased, unhealthy, or adversely affected by the concentration of the chemical because these attributes may alter accumulation of chemicals compared with healthy organisms.

b. The total concentration of the chemical in the water should be measured and should be relatively constant during the exposure period.

c. The organisms should be exposed to the chemical using a flow-through or renewal procedure.

d. The percent lipid of the tissue used to normalize the BCF_T^t should be either measured or reliably estimated to permit lipid normalization of chemical concentrations.

e. The concentrations of particulate organic carbon and dissolved organic carbon in the study water should be measured or reliably estimated.

f. Aquatic organisms used to calculate a laboratory-measured BCF_T^t should be representative of those aquatic organisms that are commonly consumed in the United States. An aquatic organism which is not commonly consumed in the United States can be used to calculate an acceptable laboratory-measured BCF_T^t provided that the organism is considered to be a reasonable surrogate for a commonly consumed organism. Information on the ecology, physiology, and biology of the organism should be reviewed when assessing whether an organism is a reasonable surrogate of a commonly consumed organism.

g. BCFs may be based on measurement of radioactivity from radiolabeled parent compounds only when the BCF is intended to include metabolites, when there is confidence that there is no interference due to metabolites of the parent compounds, or when studies are conducted to determine the extent of metabolism, thus allowing for a proper correction.

h. The calculation of the BCF_T^t should appropriately address growth dilution, which can be particularly important in affecting BCF_T^t determinations for poorly depurated chemicals.

I. Other aspects of the methodology used should be similar to those described by the American Society of Testing and Materials (ASTM, 1999) and USEPA *Ecological Effects Test Guidelines* (USEPA, 1996).

j. In addition, the magnitude of the K_{ow} and the availability of corroborating BCF data should be considered. For example, if the steady-state method is used for the BCF_T^t determination, exposure periods longer than 28 days will generally be required for highly hydrophobic chemicals to reach steady state between the water and the organism.

k. If a baseline BCF_R^{fd} derived from a laboratory-measured BCF_T^t consistently increases or decreases as the chemical concentration increases in the test solutions for the test organisms, the BCF_T^t should be selected from the test concentration(s) that would most closely correspond to the 304(a) criterion. Note: a BCF_T^t should not be calculated from a control treatment.

3. **Selecting Food Chain Multipliers.** An FCM reflects a chemical's tendency to biomagnify in the aquatic food web. Values of FCMs greater than 1.0 are indicative of biomagnification and typically apply to organic chemicals with log K_{ow} values between 4.0 and 9.0. For a given chemical, FCMs tend to be greater at higher trophic levels, although FCMs for trophic level three can be higher than those for trophic level four.

Food chain multipliers used to derive baseline BAF_R^{fd}s using Procedure #1 can be selected from model-derived or field-derived estimates.

a. **Model-Derived FCMs.** For nonionic organic chemicals appropriate for Procedure #1, EPA has calculated FCMs for various K_{ow} values and trophic levels using the bioaccumulation model of Gobas (1993). The FCMs shown in Table 5-1 were calculated using the Gobas model as the ratio of the baseline BAF_R^{fd}s for trophic levels 2, 3, and 4 to the baseline BCF_R^{fd}.

EPA recommends using the biomagnification model by Gobas (1993) to derive FCMs for nonionic organic chemicals for several reasons. First, the Gobas model includes both benthic and pelagic food chains, thereby incorporating exposure of organisms to chemicals from both the sediment and the water column. Second, the input data needed to run the model can be readily defined. Third, the predicted BAFs using the model are in agreement with field-measured BAFs for chemicals, even those with very high log K_{ow}s. Finally, the model predicts chemical residues in benthic organisms using equilibrium partitioning theory, which is consistent with EPA's equilibrium partitioning sediment guidelines (USEPA, 2000d).

The Gobas model requires input of specific data on the structure of the food chain and the water quality characteristics of the water body of interest. For calculating national BAFs, a mixed pelagic/benthic food web structure consisting of four trophic levels is assumed. Trophic level 1 is phytoplankton, trophic level 2 is zooplankton, trophic level 3 is forage fish (e.g., sculpin and smelt), and trophic level 4 are predatory fish (e.g., salmonids). Additional assumptions are made regarding the composition of the aquatic species' diets (e.g., salmonids consume 10 percent sculpin, 50 percent alewives, and 40 percent smelt), the physical parameters of the aquatic species (e.g., lipid values), and the water quality characteristics (e.g., water temperature, sediment organic carbon).

A mixed pelagic/benthic food web structure has been assumed for the purpose of calculating FCMs because it is considered to be most representative of the types of food webs that occur in aquatic ecosystems. FCMs derived using the mixed pelagic/benthic structure are also about mid-range in magnitude between a 100% pelagic and 100% benthic driven food web (see the Bioaccumulation TSD). The validity of FCMs derived using the mixed pelagic/benthic food web structure has

Table 5-1
Food-Chain Multipliers for Trophic Levels 2, 3 and 4
(Mixed Pelagic and Benthic Food Web Structure and $J_{socw} / K_{OW} = 23$)

Log K_{OW}	Trophic Level 2	Trophic Level 3	Trophic Level 4	Log K_{OW}	Trophic Level 2	Trophic Level 3	Trophic Level 4
4.0	1.00	1.23	1.07	6.6	1.00	12.9	23.8
4.1	1.00	1.29	1.09	6.7	1.00	13.2	24.4
4.2	1.00	1.36	1.13	6.8	1.00	13.3	24.7
4.3	1.00	1.45	1.17	6.9	1.00	13.3	24.7
4.4	1.00	1.56	1.23	7.0	1.00	13.2	24.3
4.5	1.00	1.70	1.32	7.1	1.00	13.1	23.6
4.6	1.00	1.87	1.44	7.2	1.00	12.8	22.5
4.7	1.00	2.08	1.60	7.3	1.00	12.5	21.2
4.8	1.00	2.33	1.82	7.4	1.00	12.0	19.5
4.9	1.00	2.64	2.12	7.5	1.00	11.5	17.6
5.0	1.00	3.00	2.51	7.6	1.00	10.8	15.5
5.1	1.00	3.43	3.02	7.7	1.00	10.1	13.3
5.2	1.00	3.93	3.68	7.8	1.00	9.31	11.2
5.3	1.00	4.50	4.49	7.9	1.00	8.46	9.11
5.4	1.00	5.14	5.48	8.0	1.00	7.60	7.23
5.5	1.00	5.85	6.65	8.1	1.00	6.73	5.58
5.6	1.00	6.60	8.01	8.2	1.00	5.88	4.19
5.7	1.00	7.40	9.54	8.3	1.00	5.07	3.07
5.8	1.00	8.21	11.2	8.4	1.00	4.33	2.20
5.9	1.00	9.01	13.0	8.5	1.00	3.65	1.54
6.0	1.00	9.79	14.9	8.6	1.00	3.05	1.06
6.1	1.00	10.5	16.7	8.7	1.00	2.52	0.721
6.2	1.00	11.2	18.5	8.8	1.00	2.08	0.483
6.3	1.00	11.7	20.1	8.9	1.00	1.70	0.320
6.4	1.00	12.2	21.6	9.0	1.00	1.38	0.210
6.5	1.00	12.6	22.8				

been evaluated in several different ecosystems including Lake Ontario, the tidally influenced Bayou D'Inde in Louisiana, the Fox River and Green Bay, Wisconsin, and the Hudson River in New York. Additional details of the validation of EPA's national default FCMs and the assumptions, uncertainties, and input parameters for the model are provided in the Bioaccumulation TSD.

Although EPA uses the FCMs in Table 5-1 to derive its national 304(a) criteria, EPA recognizes that food webs of other waterbodies might differ from the assumptions used to calculate national BAFs. In these situations, States and authorized Tribes may wish to use alternate food web structures for calculating FCMs for use in setting State or Tribal water quality criteria. Additional guidance on the use of alternate food web structures for calculating State, Tribal, or site-specific criteria is provided in the Bioaccumulation TSD.

b. **Field-Derived FCMs.** In addition to model-derived estimates of FCMs, field data may also be used to derive FCMs. Currently, the use of field-derived FCMs is the only method recommended for estimating FCMs for inorganic and organometalic chemicals because appropriate model-derived estimates are not yet available (see Section 5.6). In contrast to the model-based FCMs described previously, field-derived FCMs account for any metabolism of the chemical of concern by the aquatic organisms used to calculate the FCM.

Field-derived FCMs should be calculated using lipid-normalized concentrations of the nonionic organic chemical in appropriate predator and prey species using the following equations.

$$FCM_{TL2} = BMF_{TL2} \quad \text{(Equation 5-21)}$$

$$FCM_{TL3} = (BMF_{TL3})\,(BMF_{TL2}) \quad \text{(Equation 5-22)}$$

$$FCM_{TL4} = (BMF_{TL4})\,(BMF_{TL3})\,(BMF_{TL2}) \quad \text{(Equation 5-23)}$$

where:

FCM = Food chain multiplier for designated trophic level (TL2, TL3, or TL4)
BMF = Biomagnification factor for designated trophic level (TL2, TL3, or TL4)

The basic difference between FCMs and BMFs is that FCMs relate back to trophic level one (or trophic level two as assumed by the Gobas (1993) model), whereas BMFs always relate back to the next lowest trophic level. For nonionic organic chemicals, BMFs can be calculated from tissue residue concentrations determined in biota at a site according to the following equations.

$$BMF_{TL2} = (C_{R,\,TL2}) / (C_{R,\,TL1}) \quad \text{(Equation 5-24)}$$

$$BMF_{TL3} = (C_{R,\,TL3}) / (C_{R,\,TL2}) \quad \text{(Equation 5-25)}$$

$$BMF_{TL4} = (C_{R,\,TL4}) / (C_{R,\,TL3}) \quad \text{(Equation 5-26)}$$

where:

C_R = Lipid-normalized concentration of chemical in tissue of appropriate biota that occupy the specified trophic level (TL2, TL3, or TL4)

In addition to the acceptability guidelines pertaining to field-measured BAFs, the following procedural and quality assurance requirements apply to field-measured FCMs.

(1) Information should be available to identify the appropriate trophic levels for the aquatic organisms and appropriate predator-prey relationships for the site from which FCMs are being determined. General information on determining trophic levels of aquatic organisms can be found in USEPA 2000a,b,c.

(2) The aquatic organisms sampled from each trophic level should reflect the most important exposure pathways leading to human exposure via consumption of aquatic organisms. For higher trophic levels (e.g., 3 and 4), aquatic species should also reflect those that are commonly consumed by humans.

(3) The studies from which the FCMs are derived should contain sufficient supporting information from which to determine that tissue samples were collected and analyzed using appropriate, sensitive, accurate, and precise methods.

(4) The percent lipid should be either measured or reliably estimated for the tissue used to determine the FCM.

(5) The tissue concentrations should reflect average exposure over the approximate time required to achieve steady-state in the target species.

D. Baseline BAF_R^{fd} from a K_{ow} and FCM

The fourth method in Procedure #1 consists of using a K_{ow} and an appropriate FCM for estimating the baseline BAF_R^{fd}. In this method, the K_{ow} is assumed to be equal to the baseline BCF_R^{fd}. Numerous investigations have demonstrated a linear relationship between the logarithm of the BCF and the logarithm of the octanol-water partition coefficient (K_{ow}) for organic chemicals for fish and other aquatic organisms. Isnard and Lambert (1988) list various regression equations that illustrate this linear relationship. When the regression equations are constructed using lipid-normalized BCFs, the slopes and intercepts are not significantly different from one and zero, respectively (e.g., de Wolf, et al., 1992). The underlying assumption for the linear relationship between the BCF and K_{ow} is that the bioconcentration process can be viewed as the partitioning of a chemical between the lipid of the aquatic organisms and water and that the K_{ow} is a useful

surrogate for this partitioning process (Mackay, 1982). To account for biomagnification, Procedure #1 requires the K_{ow} value be used in conjunction with an appropriate FCM.

1. **Baseline BAF_R^{fd} Equation.** For each acceptable K_{ow} value and FCM for the chemical of concern, calculate a baseline BAF_R^{fd} using the following equation.

$$\text{Baseline } BAF_{\ell}^{fd} = (FCM) \cdot (K_{ow}) \qquad \text{(Equation 5-27)}$$

where:

Baseline BAF_R^{fd} =		BAF expressed on a freely dissolved and lipid-normalized basis for a given trophic level
FCM	=	The food chain multiplier for the appropriate trophic level obtained from Table 5-1 by linear interpolation or from appropriate field data (used with Procedure #1 only)
K_{ow}	=	Octanol-water partition coefficient

The BCF-K_{ow} relationship has been developed primarily for nonionic organic chemicals that are not readily metabolized by aquatic organisms and thus is most appropriate for poorly-metabolized nonionic organic chemicals (i.e., Procedures #1 and #3 as depicted in Figure 5-1). For poorly-metabolized nonionic organic chemicals with large log K_{ow}s (i.e., > 6), reported log BCFs are often not equal to log K_{ow}. EPA believes that this nonlinearity is primarily due to not accounting for several factors which affect the BCF determination. These factors include not basing BCFs on the freely dissolved concentration in water, not accounting for growth dilution, not assessing BCFs at steady-state, inaccuracies in measurements of uptake and elimination rate constants, and complications from the use of solvent carriers in the exposure. Application of Equation 5-27 for predicting BAFs has been conducted in several different ecosystems including Lake Ontario, the tidally influenced Bayou D'Inde in Louisiana, the Fox River and Green Bay, Wisconsin, and the Hudson River in New York. Additional detail on the validation, technical basis, assumptions, and uncertainty associated with Equation 5-27 and is provided in the Bioaccumulation TSD.

2. **FCMs and K_{ow}s**. Food chain multipliers and K_{ow} values should be selected as described previously in Procedure #1.

5.4.3.2 Selecting Final Baseline BAF_R^{fd}s

After calculating individual baseline BAF_R^{fd}s using as many of the methods in Procedure #1 as possible, the next step is to determine a final baseline BAF_R^{fd} for each trophic level from the individual baseline BAF_R^{fd}s (see Figures 5-1 and 5-2). The final baseline BAF_R^{fd} will be used in the

last step to determine the national BAF for each trophic level. The final baseline BAF_R^{fd} for each trophic level should be determined from the individual baseline BAF_R^{fd}s by considering the data preference hierarchy defined by Procedure #1 and uncertainty in the data. The data preference hierarchy for Procedure #1 is (in order of preference):

1. a baseline BAF_R^{fd} from an acceptable field-measured BAF (method 1)
2. a baseline BAF_R^{fd} predicted from an acceptable field-measured BSAF (method 2),
3. a baseline BAF_R^{fd} predicted from an acceptable BCF and FCM (method 3), or
4. a baseline BAF_R^{fd} predicted from an acceptable K_{ow} and FCM (method 4).

This data preference hierarchy reflects EPA's preference for BAFs based on field-measurements of bioaccumulation (methods 1 and 2) over those based on laboratory-measurements and/or predictions of bioaccumulation (methods 3 and 4). However, this data preference hierarchy should not be considered inflexible. Rather, it should be used as a guide for selecting the final baseline BAF_R^{fd}s when the uncertainty is similar among two or more baseline BAF_R^{fd}s derived using different methods. The following steps and guidelines should be followed for selecting the final baseline BAF_R^{fd}s using Procedure #1.

1. **Calculate Species-Mean Baseline BAF_R^{fd}s.** For each BAF method where more than one acceptable baseline BAF_R^{fd} is available for a given species, calculate a species-mean baseline BAF_R^{fd} as the geometric mean of all available individual baseline BAF_R^{fd}s. When calculating a species-mean baseline BAF_R^{fd}, individual baseline BAF_R^{fd}s should be reviewed carefully to assess the uncertainty in the BAF values. For highly hydrophobic chemicals applicable to Procedure #1, particular attention should be paid to whether sufficient spatial and temporal averaging of water and tissue concentrations was likely achieved in the BAF, BSAF, or BCF study. Highly uncertain baseline BAF_R^{fd}s should not be used. Large differences in individual baseline BAF_R^{fd}s for a given species (e.g., greater than a factor of 10) should be investigated further. In such cases, some or all of the baseline BAF_R^{fd}s for a given species might not be used. Additional discussion on evaluating acceptability of BAF values is provided in the Bioaccumulation TSD.

2. **Calculate Trophic-Level-Mean Baseline BAF_R^{fd}s.** For each BAF method where more than one acceptable species-mean baseline BAF_R^{fd} is available within a given trophic level, calculate a trophic-level-mean baseline BAF_R^{fd} as the geometric mean of acceptable species-mean baseline BAF_R^{fd}s in that trophic level. Trophic-level-mean baseline BAF_R^{fd}s should be calculated for trophic levels two, three, and four because available data on U.S. consumers of fish and shellfish indicate significant consumption of organisms in these trophic levels.

3. **Select a Final Baseline BAF_R^{fd} for Each Trophic Level.** For each trophic level, select the final baseline BAF_R^{fd} using best professional judgment by considering: (1) the data preference hierarchy shown previously, (2) the relative uncertainty in the trophic-level-mean baseline BAF_R^{fd}s derived using different methods, and (3) the weight of evidence among the four methods.

a. In general, when more than one trophic-level-mean baseline BAF_R^{fd} is available for a given trophic level, the final trophic-level-mean baseline BAF_R^{fd} should be selected from the most preferred BAF method defined by the data preference hierarchy for Procedure #1.

b. If uncertainty in a trophic-level-mean baseline BAF based on a higher tier (more preferred) method is judged to be substantially greater than a trophic-level-mean baseline BAF from a lower tier method, and the weight of evidence among the various methods suggests that a BAF value from lower tier method is likely to be more accurate, then the final baseline BAF_R^{fd} should be selected using a trophic level-mean baseline BAF_R^{fd} from a lower tier method.

c. When considering the weight of evidence among the various BAF methods, greater confidence in the final baseline BAF_R^{fd} is generally assigned when BAFs from a greater number of methods are in agreement for a given trophic level. However, lack of agreement among methods does not necessarily indicate less confidence if such disagreements can be adequately explained. For example, if the chemical of concern is metabolized by aquatic organisms represented by a BAF value, one would expect disagreement between a field-measured BAF (the highest priority data) and a predicted BAF using a K_{ow} and model-derived FCM. Thus, field-measured BAFs should generally be given the greatest weight among methods because they reflect direct measures of bioaccumulation and incorporate any metabolism which might occur in the organism and its food web.

d. The above steps should be performed for each trophic level until a final baseline BAF_R^{fd} is selected for trophic levels two, three, and four.

5.4.3.3 Calculating National BAFs

The last step in deriving a national BAF for each trophic level is to convert the final baseline BAF_R^{fd} determined in the previous step to a BAF that reflects conditions to which the national 304(a) criteria will apply (Figure 5-2). Since a baseline BAF_R^{fd} is by definition normalized by lipid content and expressed on a freely dissolved basis, it needs to be adjusted to reflect the lipid fraction of aquatic organisms commonly consumed in the U.S. and the freely dissolved fraction expected in U.S. bodies of water. Converting a final baseline BAF_R^{fd} to a national BAF requires information on: (1) the percent lipid of the aquatic organisms commonly consumed by humans, and (2) the freely dissolved fraction of the chemical of concern that would be expected in the ambient waters of interest. For each trophic level, a national BAF should be determined from a final baseline BAF_R^{fd} according to the following guidelines.

1. **National BAF Equation.** For each trophic level, calculate a national BAF using the following equation.

$$\text{National BAF}_{(TL\ n)} = [(\text{Final Baseline BAF}_{\ell}^{fd})_{TL\ n} \cdot (f_{\ell})_{TL\ n} + 1] \cdot (f_{fd}) \qquad \text{(Equation 5-28)}$$

where:

Final Baseline BAF_R^{fd}	=	Final trophic-level-mean baseline BAF expressed on a freely dissolved and lipid-normalized basis for trophic level "n"
$f_{R(TL\ n)}$	=	Lipid fraction of aquatic species consumed at trophic level "n"
f_{fd}	=	Fraction of the total chemical in water that is freely dissolved

The technical basis of Equation 5-28 is provided in the Bioaccumulation TSD. Guidance for determining each component of Equation 5-28 is provided below.

2. **Determining the Final Baseline BAF_R^{fd}.** The final trophic-level-mean baseline BAF_R^{fd}s used in this equation are those which have been determined using the guidance presented in Section 5.4.3.2 for selecting the final baseline BAF_R^{fd}s.

3. **Lipid Content of Commonly Consumed Aquatic Species.** As illustrated by Equation 5-28, the percent lipid of the aquatic species consumed by humans is needed to accurately characterize the potential exposure to a chemical from ingestion of aquatic organisms.

   a. **National Default Lipid Values.** For the purposes of calculating a national 304(a) criterion, the following national default values for lipid fraction should be used: 1.9% (for trophic level two organisms), 2.6% (for trophic level three organisms), and 3.0% (for trophic level four organisms).

      These national default values for lipid content reflect national per capita average patterns of fish consumption in the United States. Specifically, they were calculated using the consumption-weighted mean lipid content of commonly consumed fish and shellfish as identified by the USDA Continuing Survey of Food Intake by Individuals (CSFII) for 1994 through 1996. This same national survey data was used to derive national default values of fish consumption. To maintain consistency with the fish consumption assumptions, only freshwater and estuarine organisms were included in the derivation of the national default lipid values. Additional details on the technical basis, assumptions, and uncertainty in the national default values of lipid fraction are provided in the Bioaccumulation TSD.

      Although national default lipid values are used by EPA to set national 304(a) criteria, EPA encourages States and authorized Tribes to use local or regional data on lipid content of consumed aquatic species when adopting criteria into their water quality standards because local or regional consumption patterns (and lipid content) can differ from national consumption patterns. Additional guidance on

developing site-specific values of lipid content, including a database of lipid content for many commonly consumed aquatic organisms, is found in the Bioaccumulation TSD.

4. **Freely Dissolved Fraction.** The third piece of information required for deriving a national BAF is the freely dissolved fraction of the chemical of concern that is expected in waters of the United States. As noted previously, expressing BAFs on the freely dissolved concentration in water allows a common basis for averaging BAFs from several studies. However, for use in criteria development, these BAFs should be converted back to values based on the total concentration in the water to be consistent with monitored water column and effluent concentrations, which are typically based on total concentrations of chemicals in the water. This should be done by multiplying the freely dissolved baseline BAF_R^{fd} by the fraction of the freely dissolved chemical expected in water bodies of the United States where criteria are to be applied, as shown in Equation 5-29.

$$f_{fd} = \frac{1}{[1 + (POC \cdot K_{ow}) + (DOC \cdot 0.08 \cdot K_{ow})]} \qquad \text{(Equation 5-29)}$$

where:

POC = national default value for the particulate organic carbon concentration (kg/L)
DOC = national default value for the dissolved organic carbon concentration (kg/L)
K_{ow} = n-octanol water partition coefficient for the chemical

Equation 5-29 is identical to Equation 5-12, which was used to determine the freely dissolved fraction for deriving baseline BAF_R^{fd}s from field-measured BAFs. However, the POC and DOC concentrations used in Equation 5-29 reflect those values that are expected in U.S. bodies of water, not the POC and DOC values in the study water used to derive the BAF. Guidance for determining each component of Equation 5-29 follows.

a. **National Default Values of POC and DOC.** For estimating the freely dissolved fraction of the chemical of concern that is expected in U.S. water bodies, national default values of 0.5 mg/L (5×10^{-7} kg/L) for POC and 2.9 mg/L (2.9×10^{-6} kg/L) for DOC should be used. These values are 50th percentile values (medians) based on an analysis of over 110,000 DOC values and 85,000 POC values contained in EPA's STORET database from 1980 through 1999. These default values reflect a combination of values for streams, lakes and estuaries across the United States. Additional details on the technical basis, assumptions, and uncertainty in the

derivation and application of the national default values of POC and DOC are provided in the Bioaccumulation TSD.

Although national default values of POC and DOC concentrations are used by EPA to set national 304(a) criteria as described by this document, EPA encourages States and authorized Tribes to use local or regional data on POC and DOC when adopting criteria into their water quality standards. EPA encourages States and Tribes to consider local or regional data on POC and DOC because local or regional conditions may result in differences in POC or DOC concentrations compared with the values used as national defaults. Additional guidance on developing local or regional values of POC and DOC, including a database of POC and DOC values segregated by waterbody type, is found in the Bioaccumulation TSD.

b. **K_{ow}Value.** The value selected for the K_{ow} of the chemical of concern should be the same value used in earlier calculations (e.g., for calculating baseline BAF_R^{fd}s and FCMs). Guidance for selecting the K_{ow} value is found in the Bioaccumulation TSD.

5.4.4 Deriving National BAFs Using Procedure #2

This section provides guidance for calculating national BAFs for nonionic organic chemicals using Procedure #2 shown in Figure 5-1. The types of nonionic organic chemicals for which Procedure #2 is most appropriate are those that are classified as moderately to highly hydrophobic and subject to high rates of metabolism by aquatic biota (see Section 5.4.2 above). Non-aqueous contaminant exposure and subsequent biomagnification in aquatic food webs are not generally of concern for chemicals that are classified in this category. As a result, FCMs are not used in this procedure. In addition, K_{ow} -based predictions of bioconcentration are not used in this procedure since the K_{ow} /BCF relationship is primarily based on poorly metabolized chemicals. Some nonionic organic chemicals for which Procedure #2 is probably appropriate include certain PAHs which are believed to be metabolized substantially by fish (e.g., benzo[a]pyrene, phenanthrene, fluoranthene, pyrene, benzo[a]anthracene and chrysene/triphenylene; USEPA, 1980; Burkhard and Lukasewycz, 2000).

According to Procedure #2, the following three methods can be used in deriving a national BAF:

- using a BAF from an acceptable field study (i.e., a field-measured BAF) (method 1),
- predicting a BAF from an acceptable BSAF (method 2), and
- predicting a BAF from an acceptable BCF (method 3).

Each of these three methods relies on measured data for assessing bioaccumulation and therefore, includes the effects of chemical metabolism by the study organism in the BAF estimate.

The field-measured BAF and BSAF methods also incorporate any metabolism which occurs in the aquatic food web.

As shown in Figure 5-2, the next steps in deriving a national BAF after selecting the derivation procedure are: (1) calculating individual baseline BAF_R^{fd}s, (2) selecting the final baseline BAF_R^{fd}s, and (3) calculating the national BAFs. Each of these three steps is discussed separately below.

5.4.4.1 Calculating Individual Baseline BAF_R^{fd}s

As described previously in Procedure #1, calculating individual baseline BAF_R^{fd}s involves normalizing the measured BAF_T^t or BCF_T^t (which are based on the total chemical in water and tissue) by the lipid content of the study organisms and the freely dissolved fraction of the chemical in the study water. Converting measured BAF_T^t (or BCF_T^t) values to baseline BAF_R^{fd} (or BCF_R^{fd}) values is designed to account for variation in measured BAF_T^ts that is caused by differences in lipid content of study organisms and differences in the freely dissolved fraction of chemical in study waters. Therefore, baseline BAF_R^{fd}s are considered more amenable for extrapolating and averaging BAFs across different species and different study waters compared with total BAF_T^ts.

1. For each species where acceptable data are available, calculate all possible baseline BAF_R^{fd}s using each of the three methods shown above for Procedure #2.

2. Individual baseline BAF_R^{fd}s should be calculated from field-measured BAF_T^ts, field-measured BSAFs, and laboratory BCF_T^ts according to the following procedures.

A. Baseline BAF_R^{fd} from Field-Measured BAFs

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from a field-measured BAF_T^t using the guidance and equations outlined in Section 5.4.3.1(A) for determining baseline BAF_R^{fd}s from field-measured BAFs in Procedure #1.

2. Because nonionic organic chemicals applicable to Procedure #2 have relatively high rates of metabolism in aquatic organisms, they will tend to reach steady state more quickly than nonionic organic chemicals with similar K_{ow} values but which undergo little or no metabolism. Therefore, less temporal averaging of chemical concentrations would generally be required for determining field-measured BAF_T^ts with highly metabolizable chemicals compared with chemicals that are poorly metabolized by aquatic biota.

B. Baseline BAF_R^{fd} Derived from Field-measured BSAFs

1. A baseline BAF_R^{fd} should be calculated from a field-measured BSAF using the guidance and equations outlined in Section 5.4.3.1(B) for determining baseline BAF_R^{fd}s from field-measured BSAFs in Procedure #1.

C. Baseline BAF_R^{fd} from a Laboratory-Measured BCF

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from a laboratory-measured BCF_T^t using the guidance and equations outlined in Section 5.4.3.1(c) for determining baseline BAF_R^{fd}s from a laboratory-measured BCF and FCM in Procedure #1.

2. Because biomagnification is not an overriding concern for nonionic organic chemicals applicable to Procedure #2, food chain multipliers are not used in the derivation of a baseline BAF_R^{fd} from a laboratory-measured BCF_T^t .

5.4.4.2 Selecting Final Baseline BAF_R^{fd}s

After calculating individual, baseline BAF_R^{fd}s using as many of the methods in Procedure #2 as possible, the next step is to determine a final baseline BAF_R^{fd} for each trophic level from the individual baseline BAF_R^{fd}s. The final baseline BAF_R^{fd} will be used in the last step to determine the national BAF for each trophic level. A final baseline BAF_R^{fd} for each trophic level should be determined from the individual baseline BAF_R^{fd}s by considering the data preference hierarchy defined by Procedure #2 and uncertainty in the data. The data preference hierarchy for Procedure #2 is (in order of preference):

1. a baseline BAF_R^{fd} from an acceptable field-measured BAF (method 1),
2. a baseline BAF_R^{fd} from an acceptable field-measured BSAF (method 2), or
3. a baseline BAF_R^{fd} from an acceptable laboratory-measured BCF (method 3).

This data preference hierarchy reflects EPA's preference for BAFs based on field-measurements of bioaccumulation (methods 1 and 2) over those based on laboratory-measurements (method 3). However, as explained in Procedure #1, this data preference hierarchy should not be considered inflexible. Rather, it should be used as a guide for selecting the final baseline BAF_R^{fd}s when the underlying uncertainty is similar among two or more baseline BAF_R^{fd}s derived using different methods. Although biomagnification is not generally a concern for chemicals subject to Procedure #2, trophic level differences in bioaccumulation might be substantial to the extent that the rate of chemical metabolism by organisms in different trophic levels differs. For example, certain PAHs have been shown to be metabolized to a much greater extent by some fish compared with some invertebrate species (James, 1989). Therefore, final baseline BAF_R^{fd}s for chemicals applicable to Procedure #2 should be determined on a trophic-level-specific basis according to the following guidelines.

1. The final baseline BAF_R^{fd}s in Procedure #2 should be selected according to the same steps described in Procedure #1 but with the substitution of the data preference hierarchy described above for Procedure #2. Specifically, the species-mean baseline BAF_R^{fd}s, trophic-level-mean baseline BAF_R^{fd}s, and the final baseline BAF_R^{fd}s should be determined according to the guidelines presented in Procedure #1 (Section 5.4.3.2, Steps 1, 2, and 3).

5.4.4.3 Calculating the National BAFs

As described in Procedure #1, the last step in deriving national BAFs for nonionic organic chemicals is to convert the final baseline BAF_R^{fd}s determined in the previous step to BAFs which reflect conditions to which the national 304(a) criteria will apply (Figure 5-2).

1. For trophic levels two, three, and four, national BAFs should be calculated from the final baseline BAF_R^{fd}s using the same equation and procedures described previously in Procedure #1 (see Section 5.4.3.3 entitled "Calculating the National BAFs").

5.4.5 Deriving National BAFs Using Procedure #3

This section provides guidance for calculating national BAFs for nonionic organic chemicals using Procedure #3 shown in Figure 5-1. The types of nonionic organic chemicals for which Procedure #3 is most appropriate are those that are classified as low in hydrophobicity (i.e., log K_{ow} values less than 4.0) and subject to low (or unknown) rates of metabolism by aquatic biota (see Section 5.4.2 above). Non-aqueous contaminant exposure and subsequent biomagnification in aquatic food webs are not generally of concern for chemicals that are classified in this category (Fisk et al., 1998; Gobas et al., 1993; Connolly and Pedersen, 1988; Thomann, 1989). As a result, FCMs are not used in this procedure.

According to Procedure #3, the following three methods can be used in deriving a national BAF:

- using a BAF from an acceptable field study (i.e., a field-measured BAF),
- predicting a BAF from an acceptable laboratory-measured BCF, and
- predicting a BAF from an acceptable K_{ow}.

After selecting the derivation procedure, the next steps in deriving a national BAF at a given trophic level for nonionic organic chemicals are: (1) calculating individual baseline BAF_R^{fd}s, (2) selecting the final baseline BAF_R^{fd}, and (3) calculating the national BAF (Figure 5-2). Each of these three steps is discussed separately below.

5.4.5.1 Calculating Individual Baseline BAF_R^{fd}s

Calculating individual baseline BAF_R^{fd}s involves normalizing each measured BAF_T^t or BCF_T^t (which are based on the total chemical in water and tissue) by the lipid content of the study organism and the freely dissolved fraction of the chemical in the study water. For additional

discussion of the technical basis for calculating baseline BAF_R^{fd}s, see Section 5.4.3.1 in Procedure #1.

1. For each species where acceptable data are available, calculate all possible baseline BAF_R^{fd}s using each of the three methods shown above for Procedure #3.

2. An individual baseline BAF_R^{fd} should be calculated from field-measured BAF_T^ts, laboratory-measured BCF_T^ts, and K_{ow} values according to the following procedures.

A. Baseline BAF_R^{fd} from Field-Measured BAFs

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from a field-measured BAF_T^t using the guidance and equations outlined in Section 5.4.3.1(A) in Procedure #1.

2. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log K_{ow} < 4.0), nonionic organic chemicals applicable to Procedure #3 are expected to remain almost entirely in the freely dissolved form in natural waters with dissolved and particulate organic carbon concentrations typical of most field BAF studies. Therefore, the freely dissolved fraction should be assumed to be equal to 1.0, unless the concentrations of DOC and POC are very high in the field BAF study. For studies with very high DOC or POC concentrations, (e.g., about 100 mg/L or higher for DOC or 10 mg/L or higher for POC), the freely dissolved fraction may be substantially lower than 1.0 and therefore should be calculated using Equation 5-12.

3. **Temporal Averaging of Concentrations.** Also due to their low hydrophobicity, nonionic organic chemicals appropriate to Procedure #3 will also tend to reach steady state quickly compared with those chemicals to which Procedure #1 applies. Therefore, the extent of temporal averaging of tissue and water concentrations is typically much less than that required for highly hydrophobic chemicals to which Procedure #1 is applied. In addition, field studies used to calculate BAFs for these chemicals should have sampled water and tissue at similar points in time because tissue concentrations respond more rapidly to changes in water concentrations. EPA will be providing additional guidance on appropriate BAF study designs for nonionic organic chemicals (including those appropriate to Procedure #3) in its forthcoming guidance document on conducting field BAF and BSAF studies.

B. Baseline BAF_R^{fd} from a Laboratory-Measured BCF

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from a laboratory-measured BCF_T^t using the guidance and equations outlined in Section 5.4.3.1(c) of Procedure #1.

2. **Food Chain Multipliers.** Because biomagnification is not an overriding concern for the minimally hydrophobic chemicals applicable to Procedure #3, FCMs are not used in the derivation of a baseline BAF_R^{fd} from a laboratory-measured BCF_T^t.

3. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log K_{ow} < 4.0), nonionic organic chemicals to which Procedure #3 is applied are expected to remain almost entirely in the freely dissolved form in waters containing dissolved and particulate organic carbon concentrations typical of laboratory BCF studies. Therefore, the freely dissolved fraction should usually be assumed equal to 1.0. The freely dissolved fraction will be substantially less than 1.0 only in situations where unusually high concentrations of DOC and POC are present in the laboratory BCF study (e.g., above about 100 mg/L for DOC or about 10 mg/L for POC). In this situation, the freely dissolved fraction should be calculated according to Equation 5-12.

C. Baseline BAF_R^{fd} from a K_{ow}

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from an acceptable K_{ow} using the guidance and equations outlined in Section 5.4.3.1(D) in Procedure #1.

2. Because biomagnification is not an overriding concern for nonionic organic chemicals with low hydrophobicity (i.e., log K_{ow} < 4.0), food chain multipliers are not used in Procedure #3 for deriving the baseline BAF_R^{fd} from a K_{ow}.

5.4.5.2 Selecting Final Baseline BAF_R^{fd}s

After calculating individual baseline BAF_R^{fd}s using as many of the methods in Procedure #3 as possible, the next step is to determine a final baseline BAF_R^{fd} for each trophic level from the individual baseline BAF_R^{fd}s (Figure 5-2). The final baseline BAF_R^{fd} will be used in the last step to determine the national BAF for each trophic level. The final baseline BAF_R^{fd} for each trophic level should be determined from the individual baseline BAF_R^{fd}s by considering the data preference hierarchy defined by Procedure #3 and uncertainty in the data. The data preference hierarchy for Procedure #3 is (in order of preference):

1. a baseline BAF_R^{fd} from an acceptable field-measured BAF or laboratory-measured BCF, or
2. a baseline BAF_R^{fd} predicted from an acceptable K_{ow} value.

This data preference hierarchy reflects EPA's preference for BAFs that are based on measured data (field-measured BAFs and laboratory-measured BCFs) over BAFs based on predictive methods (K_{ow}). This data preference hierarchy should be used as a guide for selecting the final baseline BAF_R^{fd}s when the uncertainty is similar among two or more baseline BAF_R^{fd}s derived using different methods. Since bioaccumulation via dietary uptake and subsequent biomagnification generally are not of concern for chemicals subject to Procedure #3, field-

measured BAFs and laboratory-measured BCFs are considered equally in determining the national BAF.

Final baseline BAF_R^{fd}s should be selected for each trophic level using the following steps and guidelines.

1. **Calculate Species-Mean Baseline BAF_R^{fd}s.** For each BAF method (i.e., field-measured BAF, BAF from a lab-measured BCF, or BAF from a K_{ow}) where more than one acceptable baseline BAF_R^{fd} is available for a given species, calculate a species-mean baseline BAF_R^{fd} according to the guidance described previously in Procedure #1.

2. **Calculate Trophic-Level-Mean Baseline BAF_R^{fd}s.** For each BAF method where more than one acceptable species-mean baseline BAF_R^{fd} is available within a given trophic level, calculate the trophic-level-mean baseline BAF_R^{fd} as the geometric mean of acceptable species-mean baseline BAF_R^{fd}s in that trophic level.

3. **Select a Final Baseline BAF_R^{fd} for Each Trophic Level.** For each trophic level, select the final baseline BAF_R^{fd} using best professional judgment by considering: (1) the data preference hierarchy, (2) the relative uncertainties among trophic-level-mean baseline BAF_R^{fd}s derived using different methods, and (3) the weight of evidence among the three methods.

    a. In general, when more than one trophic-level-mean baseline BAF_R^{fd} is available within a given trophic level, the final baseline BAF_R^{fd} should be selected from the most preferred BAF method defined by the data preference hierarchy for Procedure #3. Within the first data preference tier, field-measured BAFs and laboratory-measured BCFs are considered equally desirable for deriving a final trophic-level-mean baseline BAF_R^{fd} using Procedure #3. If a trophic-level-mean baseline BAF_R^{fd} is available from both a field-measured BAF and a laboratory-measured BCF, the final baseline BAF_R^{fd} should be selected using the trophic-level-mean baseline BAF_R^{fd} or BCF_R^{fd} with the least overall uncertainty.

    b. If uncertainty in a trophic-level-mean baseline BAF_R^{fd} based on a higher tier (more preferred) method is judged to be substantially greater than a trophic-level-mean baseline BAF_R^{fd} from a lower tier method, then the final baseline BAF_R^{fd} should be selected using a trophic-level-mean baseline BAF_R^{fd} from a lower tier method.

    c. The above steps should be performed for each trophic level until a final baseline BAF_R^{fd} is selected for trophic level two, three, and four.

5.4.5.3 Calculating the National BAFs

As described in Procedure #1, the last step in deriving a national BAF for a given trophic level for nonionic organic chemicals is to convert the final baseline BAF_R^{fd} determined in the

previous step to a BAF that reflect conditions to which the national 304(a) criterion will apply (Figure 5-2). Each national BAF should be determined from a final baseline BAF_R^{fd} according to the following guidelines.

1. **National BAF Equation.** Except where noted below, national BAFs for trophic levels two, three, and four should be calculated from the final, trophic-level-mean baseline BAF_R^{fd}s using Equation 5-28 and associated guidance described in Procedure #1 (see Section 5.4.3.3).

2. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log K_{ow} < 4.0), a freely dissolved fraction of 1.0 should be assumed for calculating national BAFs for nonionic organic chemicals using Procedure #3. A freely dissolved fraction of 1.0 should be assumed because at a log K_{ow} of less than 4.0, nonionic organic chemicals are expected to remain over 99 percent in the freely dissolved form at POC and DOC concentrations corresponding to national default values for U.S. bodies of water (i.e., 0.5 mg/L and 2.9 mg/L, respectively).

5.4.6 Deriving National BAFs Using Procedure #4

This section provides guidance for calculating national BAFs for nonionic organic chemicals using Procedure #4 shown in Figure 5-1. The types of nonionic organic chemicals for which Procedure #4 is most appropriate are those that are classified as having low hydrophobicity and subject to high rates of metabolism by aquatic biota (see Section 5.4.2 above). Non-aqueous contaminant exposure and subsequent biomagnification in aquatic food webs are not generally of concern for chemicals that are classified in this category. As a result, FCMs are not used in this procedure. In addition, K_{ow} -based predictions of bioconcentration are not used in this procedure since the K_{ow} /BCF relationship is primarily based on poorly metabolized chemicals. One example of a nonionic organic chemical for which Procedure #4 appears appropriate is butyl benzyl phthalate in fish. Using radiolabeling techniques with confirmation by chromatographic analysis, Carr et al. (1997) present evidence that indicates butyl benzyl phthalate is extensively metabolized in sunfish. Carr et al. (1997) also report measured BCFs (and subsequently lipid-normalized BCFs) which are substantially below predicted BCFs based on log $K_{ow.}$ In a study of chlorinated anilines (which would be essentially un-ionized at ambient pH), de Wolf et al. (1992) reported measured BCFs substantially lower than those predicted based on K_{ow}. The authors suggested that biotransformation (metabolism) involving the amine (NH_2) was responsible for the lower measured BCFs.

According to Procedure #4, the following two methods can be used in deriving a national BAF:

- using a BAF from an acceptable field study (i.e., a field-measured BAF), and
- predicting a BAF from an acceptable BCF.

After selecting the derivation procedure, the next steps in deriving a national BAF for a given trophic level for nonionic organic chemicals are: (1) calculating individual baseline BAF_R^{fd}s, (2) selecting the final baseline BAF_R^{fd}, and (3) calculating the national BAF (Figure 5-2). Each of these three steps is discussed separately below.

5.4.6.1 Calculating Individual Baseline BAF_R^{fd}s

Calculating individual baseline BAF_R^{fd}s involves normalizing the measured BAF_T^t or BCF_T^t (which are based on the total chemical in water and tissue) by the lipid content of the study organism and the freely dissolved fraction of the chemical in the study water. For additional discussion of the technical basis for calculating baseline BAF_R^{fd}s, see Section 5.4.3.1 in Procedure #1.

1. For each species where acceptable data are available, calculate all possible baseline BAF_R^{fd}s using each of the two methods shown above for Procedure #4.

2. Individual baseline BAF_R^{fd}s should be calculated from field-measured BAF_T^ts and laboratory-measured BCF_T^ts according to the following procedures.

A. Baseline BAF_R^{fd} from Field-Measured BAFs

1. A baseline BAF_R^{fd} should be calculated from a field-measured BAF_T^t using the guidance and equations outlined in Section 5.4.3.1(A) in Procedure #1.

2. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log $K_{ow} < 4.0$), nonionic organic chemicals applicable to Procedure #4 are expected to remain almost entirely in the freely dissolved form in natural waters with dissolved and particulate organic carbon concentrations typical of most field BAF studies. Therefore, the freely dissolved fraction should be assumed equal to 1.0 unless the concentrations of DOC and POC are very high in the field BAF study. For studies with very high DOC or POC concentrations, (e.g., about 100 mg/L or higher for DOC or 10 mg/L or higher for POC), the freely dissolved fraction may be substantially lower than 1.0 and therefore should be calculated using Equation 5-12.

3. **Temporal Averaging of Concentrations.** Also due to their low hydrophobicity, nonionic organic chemicals appropriate to Procedure #4 will also tend to reach steady-state quickly compared with those chemicals to which Procedure #1 applies. Therefore, the extent of temporal averaging of tissue and water concentrations is typically much less than that required for highly hydrophobic chemicals to which Procedure #1 is applied. In addition, field studies used to calculate BAFs for these chemicals should have sampled water and tissue at similar points in time because tissue concentrations should respond rapidly to changes in water concentrations. EPA will be providing additional guidance on appropriate BAF study designs for nonionic organic chemicals (including those

appropriate to Procedure #4) in its forthcoming guidance document on conducting field BAF and BSAF studies.

B. Baseline BAF_R^{fd} from a Laboratory-Measured BCF

1. Except where noted below, a baseline BAF_R^{fd} should be calculated from a laboratory-measured BCF_T^t using the guidance and equations outlined in Section 5.4.3.1(c) of Procedure #1.

2. **Food Chain Multipliers.** Because biomagnification is not an important concern for the minimally hydrophobic chemicals applicable to Procedure #4, FCMs are not used in the derivation of a baseline BAF_R^{fd} from a laboratory-measured BCF_T^t.

3. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log K_{ow} < 4.0), nonionic organic chemicals to which Procedure #4 is applied are expected to remain almost entirely in the freely dissolved form in waters containing dissolved and particulate organic carbon concentrations typical of laboratory BCF studies. Therefore, the freely dissolved fraction should usually be assumed to be equal to 1.0. The freely dissolved fraction will be substantially less than 1.0 only in situations where unusually high concentrations of DOC and POC are present in the lab BCF study (e.g., above about 100 mg/L for DOC or about 10 mg/L for POC). In this situation, the freely dissolved fraction should be calculated according to Equation 5-12.

5.4.6.2 Selecting the Final Baseline BAF_R^{fd}s

After calculating individual baseline BAF_R^{fd}s using as many of the methods in Procedure #4 as possible, the next step is to determine a final baseline BAF_R^{fd} for a given trophic level from the individual baseline BAF_R^{fd}s (Figure 5-2). The final baseline BAF_R^{fd} will be used in the last step to determine the national BAF for each trophic level. A final baseline BAF_R^{fd} should be determined for each trophic level from the individual baseline BAF_R^{fd}s by considering the data preference hierarchy defined by Procedure #4 and uncertainty in the data. The data preference hierarchy for Procedure #4 is:

1. a baseline BAF_R^{fd} from an acceptable field-measured BAF or predicted from an acceptable laboratory-measured BCF.

Since bioaccumulation via dietary uptake and subsequent biomagnification generally are not of concern for chemicals subject to Procedure #4, field-measured BAFs and laboratory-measured BCFs are considered equally in determining the national BAF.

Final baseline BAF_R^{fd}s should be selected for each trophic level using the following steps and guidelines.

1. **Calculate Species-Mean Baseline BAF_R^{fd}s.** For each BAF method (i.e., field-measured BAF or a BAF from a lab-measured BCF) where more than one acceptable baseline BAF_R^{fd} is available for a given species, calculate a species-mean baseline BAF_R^{fd} according to the guidance described previously in Procedure #1.

2. **Calculate Trophic-Level-Mean Baseline BAF_R^{fd}s.** For each BAF method where more than one acceptable species-mean baseline BAF_R^{fd} is available within a given trophic level, calculate the trophic-level-mean baseline BAF_R^{fd} as the geometric mean of acceptable species-mean baseline BAF_R^{fd}s for that trophic level.

3. **Select a Final Baseline BAF_R^{fd} for Each Trophic Level.** For each trophic level, select the final baseline BAF_R^{fd} using best professional judgment by considering: (1) the data preference hierarchy, and (2) the relative uncertainties among trophic-level-mean BAFs derived using different methods.

    a. As discussed above, field-measured BAFs and laboratory-measured BCFs are considered equally desirable for deriving a final trophic-level-mean baseline BAF_R^{fd} using Procedure #4. If a trophic-level-mean baseline BAF_R^{fd} is available from both a field-measured BAF and a laboratory-measured BCF, the final baseline BAF_R^{fd} should be selected using the trophic-level-mean baseline BAF_R^{fd} or BCF_R^{fd} with the least overall uncertainty.

    b. The above steps should be performed for each trophic level until a final baseline BAF_R^{fd} is selected for trophic levels two, three, and four.

5.4.6.3 Calculating National BAFs

As described in Procedure #1, the last step in deriving a national BAF for a given trophic level for nonionic organic chemicals is to convert the final baseline BAF_R^{fd} determined in the previous step to a BAF that reflects conditions to which the national 304(a) criterion will apply (Figure 5-2). Each national BAF should be determined from a final baseline BAF_R^{fd} according to the following guidelines.

1. **National BAF Equation.** Except where noted below, national BAFs for trophic-levels two, three, and four should be calculated from the final, trophic-level-mean baseline BAF_R^{fd}s using the same equation and procedures described previously in Procedure #1 (see Section 5.4.3.3 in Procedure #1).

2. **Freely Dissolved Fraction**. Due to their low hydrophobicity (i.e., log K_{ow} < 4.0), a freely dissolved fraction of 1.0 should be assumed for calculating national BAFs for nonionic organic chemicals using Procedure #4. A freely dissolved fraction of 1.0 should be assumed because at a log K_{ow} value of less than 4.0, nonionic organic chemicals are expected to remain over 99 percent in the freely dissolved form at POC and DOC

concentrations corresponding to national default values for U.S. bodies of water (i.e., 0.5 mg/L and 2.9 mg/L, respectively).

5.5 NATIONAL BIOACCUMULATION FACTORS FOR IONIC ORGANIC CHEMICALS

This section contains guidelines for deriving national BAFs for ionic organic chemicals (i.e., organic chemicals which undergo significant ionization in water). As defined in Section 5.3.5, ionic organic chemicals contain functional groups which can either readily donate protons (e.g., organic acids with hydroxyl, carboxylic, and sulfonic groups) or readily accept protons (e.g., organic bases with amino and aromatic heterocyclic nitrogen groups). Some examples of ionic organic compounds include:

- chlorinated phenols (e.g., 2,4,6-trichlorophenol, pentachlorophenol),
- chlorinated phenoxyalkanoic acids (e.g., 2,4-dichlorophenoxyacetic acid [2,4-D]),
- nitrophenols (e.g., 2-nitrophenol, 2,4,6-trinitrophenol),
- cresols (e.g., 2,4-dinitro-*o*-cresol [DNOC]),
- pyridines (e.g., 2,4-dimethypyidine),
- aliphatic and aromatic amines (e.g., trimethylamine, aniline), and
- linear alkylbenzenesulfonate (LAS) surfactants.

Ionic organic chemicals are considered separately for deriving national BAFs because the anionic or cationic species of these chemicals behave much differently in the aquatic environment compared with their neutral (un-ionized) counterparts. The neutral species of ionic organic chemicals are thought to behave in a similar manner as nonionic organic compounds (e.g., partitioning to lipids and organic carbon as a function of hydrophobicity). However, the ionized (cationic, anionic) species exhibit a considerably more complex behavior involving multiple environmental partitioning mechanisms (e.g., ion exchange, electrostatic, and hydrophobic interactions) and a dependency on pH and other factors including ionic strength and ionic composition (Jafvert et al., 1990; Jafvert 1990; Schwarzenbach, et al., 1993). As a consequence, methods to predict the environmental partitioning of organic cations and anions are less developed and validated compared with methods for nonionic organic chemicals (Spacie, 1994; Suffet et al., 1994).

Given the current limitations in the state of the science for predicting the partitioning and bioaccumulation of the ionized species of ionic organic chemicals, procedures for deriving national BAFs for these chemicals differ depending on the extent to which the fraction of the total chemical is likely to be represented by the ionized (cationic, anionic) species in U.S. surface waters. When a significant fraction of the total chemical concentration is expected to be present as the ionized species in water, procedures for deriving the national BAF rely on empirical (measured) methods (i.e., Procedures #5 and 6 in Section 5.6). When an insignificant fraction of the total chemical is expected to be present as the ionized species (i.e., the chemical exists essentially in the neutral form), procedures for deriving the national BAF will follow those

established for nonionic organic chemicals (e.g., Procedures #1 through #4 in Section 5.4). The following guidelines apply for assessing the occurrence of cationic and anionic forms at typical environmental pH ranges.

1. For the ionic organic chemical of concern, the dissociation constant, pK_a, should be compared to the range of pH values expected in fresh and estuarine waters of the U.S. At pH equal to the pK_a, 50% of the organic acid or base is expected to be present in the ionized species. The pH values for U.S. fresh and estuarine waters typically range between 6 and 9, although somewhat higher and lower values can occur in some bodies of water (e.g., acidic bogs and lakes, highly alkaline and eutrophic systems, etc.).

2. For organic acids, the chemical will exist almost entirely in its un-ionized form when pH is about 2 or more units below the pK_a. For organic bases, the chemical will exist almost entirely in its un-ionized form when pH is about 2 or more units above the pK_a. In these cases, the aqueous behavior of the chemical would be expected to be similar to nonionic organic chemicals. Therefore, national BAF should usually be derived using Procedures #1 through #4 in Section 5.4.

3. When pH is greater than the pK_a minus 2 for organic acids (or less than the pKa plus 2 for organic bases), the fraction of the total chemical that is expected to exist in its ionized form can become significant (i.e., $\geq$1% in the ionized). In these cases, the national BAF should usually be derived using Procedures #5 and #6 in Section 5.6.

4. In general, most organic acids (e.g., pentachlorophenol and silvex), exist primarily in the ionized form in ambient waters because their pK_a's (4.75 and 3.07, respectively) are much smaller than the pH of the ambient waters. Conversely, most organic bases, (e.g., aniline) exist mostly in the un-ionized form in ambient waters because their pK_a's (4.63 for aniline) are much smaller than the pH of the ambient waters.

5. The above guidelines are intended to be a general guide for deriving national BAFs for ionic organic chemicals, not an inflexible rule. Modifications to these guidelines should be considered on a case-by-case basis, particularly when such modifications are strongly supported by measured bioaccumulation or bioconcentration data. For example, initial models have been developed for predicting the solid and organic-phase partitioning of certain organic acids (e.g., Jafvert 1990, Jafvert et al., 1990). As these or other models become more fully developed and appropriately validated in the future, they should be considered in the development of national BAFs. In addition, since pH is a controlling factor for dissociation and subsequent partitioning of ionic organic chemicals, consideration should be given to expressing BAFs or BCFs as a function of pH (or other factors) where sufficient data exist to reliably establish such relationships.

5.6 NATIONAL BIOACCUMULATION FACTORS FOR INORGANIC AND ORGANOMETALLIC CHEMICALS

This section contains guidelines for deriving national BAFs for inorganic and organometallic chemicals as defined in Section 5.3.5. The derivation of BAFs for inorganic and organometallic chemicals differs in several ways from procedures for nonionic organic chemicals. First, lipid normalization of chemical concentrations in tissues does not generally apply for inorganic and organometallic chemicals. Thus, BAFs and BCFs cannot be extrapolated from one tissue to another based on lipid-normalized concentrations as is done for nonionic organic chemicals. Second, the bioavailability of inorganics and organometallics in water tends to be chemical-specific and thus, the techniques for expressing concentrations of nonionic organic chemicals based on the freely dissolved form do not generally apply. Third, at the present time there are no generic bioaccumulation models that can be used to predict BAFs for inorganic and organometallic chemicals as a whole, unlike the existence of K_{ow}-based models for nonionic organic chemicals. While some chemical-specific bioaccumulation models have been developed for inorganic and organometallic chemicals (e.g., Mercury Cycling Model by Hudson et. al, 1994), those models currently tend to require site-specific data for input to the model and are restricted to site-specific applications. As the models become more fully developed and validated in the future, they should be considered on a case-by-case basis in conjunction with the following procedures for deriving national BAFs.

5.6.1 Selecting the BAF Derivation Procedure

As shown in Figure 5-1, national BAFs can be derived using two procedures for inorganic and organometallic chemicals (Procedures #5 and #6). The choice of the BAF derivation procedure depends on whether or not the chemical undergoes biomagnification in aquatic food webs.

1. For many inorganic and organometallic chemicals, biomagnification does not occur and the BCF will be equal to the BAF. For these types of chemicals, Procedure #5 should be used to derive the national BAF. Procedure #5 considers BAFs and BCFs to be of equal value in determining the national BAF and does not require the use of FCMs with BCF measurements. Guidance for deriving BAFs using Procedure #5 is provided in Section 5.6.3.

2. For some inorganic and organometallic chemicals (e.g., methylmercury), biomagnification does occur and Procedure #6 should be used to determine the national BAF. Procedure #6 gives general preference to the use of field-measured BAFs over laboratory-measured BCFs and requires FCMs to be used with BCF measurements for predicting BAFs. Guidance for deriving BAFs using Procedure #6 is provided in Section 5.6.4.

3. Determining whether or not biomagnification occurs for inorganic and organometallic chemicals requires chemical-specific data on measured concentrations of the chemical in aquatic organisms and their prey. Concentrations in aquatic organisms that increase

substantially at successive trophic levels of a food web suggest that biomagnification is occurring. Concentrations in aquatic organisms that remain about the same or decrease at successive trophic levels of a food web suggest that biomagnification is not occurring. When comparing tissue concentrations for assessing biomagnification, care should be taken to ensure that the aquatic organisms chosen actually represent functional predator-prey relationships and that all major prey species are considered in the comparisons.

5.6.2 Bioavailability

The chemical-specific nature of inorganic and organometallic bioavailability is likely due in part to chemical-specific differences in several factors which affect bioavailability and bioaccumulation. These factors include differences in the mechanisms for chemical uptake by aquatic organisms (e.g., passive diffusion, facilitated transport, active transport), differences in sorption affinities to biotic and abiotic ligands, and differences in chemical speciation in water. Some inorganic and organometallic chemicals exist in multiple forms and valence states in aquatic ecosystems that can differ in their bioavailability to aquatic organisms and undergo conversions between forms. For example, selenium can exist in various forms in aquatic ecosystems, including inorganic selenite($^{+4}$) and selenate($^{+6}$) oxyanions, elemental selenium (^{0}) under reducing conditions (primarily in sediments), and organoselenium compounds of selenide ($^{-2}$). Dominant forms of mercury in natural, oxic waters include inorganic ($^{+2}$) mercury compounds and methylmercury; the latter is generally considered to be substantially more bioavailable than inorganic mercury compounds to higher trophic level organisms. Although a generic analogue to the "freely dissolved" conversion for nonionic organic chemicals does not presently exist for inorganic and organometallic chemicals as a whole, the occurrence and bioavailability of different forms of these chemicals should be carefully considered when deriving national BAFs.

1. If data indicate that: (1) a particular form (or multiple forms) of the chemical of concern largely governs its bioavailability to target aquatic organisms, and (2) BAFs are more reliable when derived using the bioavailable form(s) compared with using other form(s) of the chemical of concern, then BAFs and BCFs should be based on the appropriate bioavailable form(s).

2. Because different forms of many inorganic and organometallic chemicals may interconvert once released to the aquatic environment, regulatory and mass balance considerations typically require an accounting of the total concentration in water. In these cases, sufficient data should be available to enable conversion between total concentrations and the other (presumably more bioavailable) forms in water.

5.6.3 Deriving BAFs Using Procedure #5

This section contains guidance for calculating national BAFs for inorganic and organometallic chemicals using Procedure #5 as shown in Figure 5-1. The types of inorganic and

organometallic chemicals for which Procedure #5 is appropriate are those that are not likely to biomagnify in aquatic food webs (see Section 5.1 above). In Procedure #5, two methods are available to derive the national BAF for a given trophic level:

- using a BAF from an acceptable field study (i.e., field-measured BAF), or
- predicting a BAF from an acceptable laboratory-measured BCF.

Individual BAFs should be determined from field-measured BAFs or laboratory-measured BCFs according to the following guidelines.

5.6.3.1 Determining Field-Measured BAFs

1. Except where noted below, field-measured BAFs should be determined using the guidance provided in Section 5.4.3.1(A) of Procedure #1.

2. As described previously, conversion of field-measured BAFs to baseline BAF_R^{fd}s based on lipid-normalized and freely-dissolved concentrations does not apply for inorganic and organometallic chemicals. Therefore, the guidance and equations provided in Procedure #1 which pertain to converting field-measured BAFs to baseline BAF_R^{fd}s and subsequently to national BAFs do not generally apply to inorganic chemicals. As discussed in Section 5.6.2 above, an analogous procedure in concept might be required for converting total BAFs to BAFs based on the most bioavailable form(s) for some inorganic and organometallic chemicals of concern. Such procedures should be applied on a chemical-specific basis.

3. BAFs should be expressed on a wet-weight basis; BAFs reported on a dry-weight basis can be used only if they are converted to a wet-weight basis using a conversion factor that is measured or reliably estimated for the tissue used in the determination of the BAF.

4. BAFs should be based on concentrations in the edible tissue(s) of the biota unless it is demonstrated that whole-body BAFs are similar to edible tissue BAFs. For some finfish and shellfish species, whole body is considered to be the edible tissue.

5. The concentrations of an inorganic or organometallic chemical in a bioaccumulation study should be greater than normal background levels and greater than levels required for normal nutrition of the test species if the chemical is a micronutrient, but below levels that adversely affect the species. Bioaccumulation of an inorganic or organometallic chemical that is essential to the nutrition of aquatic organisms might be overestimated if concentrations are at or below normal background levels due to selective accumulation by the organisms to meet their nutritional requirements.

5.6.3.2 Determining Laboratory-Measured BCFs

1. Except where noted below, BAFs should be predicted from laboratory-measured BCFs using the guidance provided in Section 5.4.3.1(c) of Procedure #1.

2. As described previously, conversion of laboratory-measured BCFs to baseline BCF_R^{fd}s based on lipid-normalized and freely dissolved concentrations does not apply for inorganic and organometallic chemicals. Therefore, the guidance and equations provided in Procedure #1 which pertain to converting laboratory-measured BCFs to baseline BCF_R^{fd}s and subsequently to national BCFs do not generally apply to inorganic and organometallic chemicals. As discussed in Section 5.6.2 above, an analogous procedure in concept might be required for converting total BCFs to BCFs based on the most bioavailable form(s) of some inorganic and organometallic chemicals of concern. Such procedures should be applied on a chemical-specific basis. In addition, the use of FCMs with BCFs does not apply to chemicals applicable to Procedure #5.

3. BCFs should be expressed on a wet-weight basis; BCFs reported on a dry-weight basis can be used only if they are converted to a wet-weight basis using a conversion factor that is measured or reliably estimated for the tissue used in the determination of the BCF.

4. BCFs should be based on concentrations in the edible tissue(s) of the biota unless it is demonstrated that whole-body BCFs are similar to edible tissue BCFs. For some finfish and shellfish species, whole body is considered to be the edible tissue.

5. The concentrations of an inorganic or organometallic chemical in a bioconcentration test should be greater than normal background levels and greater than levels required for normal nutrition of the test species if the chemical is a micronutrient, but below levels that adversely affect the species. Bioaccumulation of an inorganic or organometallic chemical that is essential to the nutrition of aquatic organisms might be overestimated if concentrations are at or below normal background levels due to selective accumulation by the organisms to meet their nutritional requirements.

5.6.3.3 Determining the National BAFs

After calculating individual BAFs using as many of the methods in Procedure #5 as possible, the next step is to determine national BAFs for each trophic level from the individual BAFs. The national BAFs will be used to determine the national 304(a) criteria. The national BAFs should be determined from the individual BAFs by considering the data preference hierarchy defined for Procedure #5 and uncertainty in the data. The data preference hierarchy for Procedure #5 is:

1. a BAF from an acceptable field-measured BAF or predicted from an acceptable laboratory-measured BCF.

Since bioaccumulation via dietary uptake and subsequent biomagnification are not of concern for chemicals subject to Procedure #5, field-measured BAFs and laboratory-measured BCFs are considered equally in determining the national BAFs. The national BAFs should be selected for each trophic level using the following steps and guidelines.

1. **Calculate Species-Mean BAFs.** For each BAF method where more than one acceptable field-measured BAF (or a BAF predicted from a BCF) is available for a given species, calculate the species-mean BAF as the geometric mean of all acceptable individual measured or BCF-predicted BAFs. When calculating species-mean BAFs, individual measured or BCF-predicted BAFs should be reviewed carefully to assess uncertainties in the BAF values. Highly uncertain BAFs should not be used. Large differences in individual BAFs for a given species (e.g., greater than a factor of 10) should be investigated further and in such cases, some or all of the BAFs for a given species might not be used. Additional discussion on evaluating the acceptability of BAF and BCF values is provided in the Bioaccumulation TSD.

2. **Calculate Trophic-Level-Mean BAFs.** For each BAF method where more than one acceptable species-mean BAF is available within a given trophic level, calculate the trophic-level-mean BAF as the geometric mean of acceptable species-mean BAFs in that trophic level. Trophic-level-mean BAFs should be calculated for trophic levels two, three and four because available data on U.S. consumers of fish and shellfish indicate significant consumption of organisms in these trophic levels.

3. **Select a Final National BAF for Each Trophic Level.** For each trophic level, select the final national BAF using best professional judgment by considering: (1) the data preference hierarchy in Procedure #5, and (2) the relative uncertainties among trophic level-mean BAFs derived using different methods.

    a. As discussed above, field-measured BAFs and laboratory-measured BCFs are considered equally desirable for deriving a final national BAF using Procedure #5. If a trophic-level-mean BAF is available from both a field-measured BAF and a laboratory-measured BCF, the final national BAF should be selected using the trophic-level-mean BAF with the least overall uncertainty.

    b. The above steps should be performed for each trophic level until a national BAF is selected for trophic levels two, three, and four.

5.6.4 Deriving BAFs Using Procedure #6

This section contains guidance for calculating national BAFs for inorganic and organometallic chemicals using Procedure #6 as shown in Figure 5-1. The types of inorganic and organometallic chemicals for which Procedure #6 is appropriate are those that are considered likely to biomagnify in aquatic food webs (see Section 5.6.1 above). Methylmercury is an

example of an organometallic chemical to which Procedure #6 applies. In Procedure #6, two methods are available to derive the national BAF:

- using a BAF from an acceptable field study (i.e., field-measured BAF), or
- predicting a BAF from an acceptable laboratory-measured BCF and a FCM.

Individual BAFs should be determined from field-measured BAFs or laboratory-measured BCFs and FCMs according to the following guidelines.

5.6.4.1 Determining Field-Measured BAFs

1. Field-measured BAFs should be determined using the guidance provided in Section 5.6.3.1 of Procedure #5.

5.6.4.2 Determining Laboratory-Measured BCFs

1. Except where noted below, BAFs should be predicted from laboratory-measured BCFs using the guidance provided in Section 5.6.3.2 of Procedure #5.

2. Because biomagnification is of concern for chemicals applicable to Procedure #6, BAFs should be predicted from laboratory-measured BCF using FCMs. Currently, there are no generic models from which to predict FCMs for inorganic or organometallic chemicals. Therefore, FCMs should be determined using field data as described in the section entitled: "Field-Derived FCMs" in Section 5.4.3.1(c) of Procedure #1. Unlike nonionic organic chemicals, field-derived FCMs for inorganic and organometallic chemicals are not based on lipid-normalized concentrations in tissues. For calculating FCMs for inorganic and organometallic chemicals, concentrations in tissues should be based on the consistent use of either wet-weight or dry-weight concentrations in edible tissues. FCMs should be derived for trophic levels two, three, and four.

5.6.4.3 Determining the National BAF

After calculating individual BAFs using as many of the methods in Procedure #6 as possible, the next step is to determine national BAFs for each trophic level from the individual BAFs. The national BAFs will be used to determine the national 304(a) criteria. The national BAFs should be determined from the individual BAFs by considering the data preference hierarchy defined for Procedure #6 and uncertainty in the data. The data preference hierarchy for Procedure #6 is (in order of preference):

1. a BAF from an acceptable field-measured BAF, or
2. a predicted BAF from an acceptable laboratory-measured BCF and FCM.

This data preference hierarchy reflects EPA's preference for field-measured BAFs over BAFs predicted from a laboratory-measured BCF and FCM, because field-measured BAFs are

direct measures of bioaccumulation and biomagnification in aquatic food webs. BAFs predicted from laboratory-measured BCFs and FCMs indirectly account for biomagnification through the use of the FCM. For each trophic level, the national BAFs should be determined using the following steps and guidelines.

1. **Calculate Species-Mean BAFs.** For each BAF method where more than one acceptable field-measured BAF or BAF predicted using a BCF and FCM is available, calculate a species-mean BAF according to the guidance described previously in Procedure #5.

2. **Calculate Trophic Level-Mean BAFs.** For each BAF method where more than one acceptable species-mean BAF is available within a given trophic level, calculate the trophic level-mean BAF according to guidance described previously in Procedure #5.

3. **Select a Final National BAF for Each Trophic Level.** For each trophic level, select the final national BAF using best professional judgment by considering: (1) the data preference hierarchy in Procedure #6, and (2) the relative uncertainties among trophic level-mean BAFs derived using different methods.

    a. When a trophic-level mean BAF is available using both methods for a given trophic level (i.e., a field-measured BAF and a BAF predicted from a BCF and FCM), the national BAF should usually be selected using the field-measured BAF which is the preferred BAF method in the data preference hierarchy in Procedure #6.

    b. If uncertainty in the trophic-level mean BAF derived using field-measured BAFs is considered to be substantially greater than a trophic-level mean BAF derived using a BCF and FCM, the national BAF for that trophic level should be selected from the second tier (BCF @ FCM) method.

    c. The above steps should be performed for each trophic level until a national BAF is selected for trophic levels two, three, and four.

5.7 REFERENCES

ASTM (American Society of Testing and Materials). 1999. Standard practice for conducting bioconcentration tests with fishes and saltwater bivalve molluscs. Designation E 1022 - 94. In: *Annual Book of ASTM standards*. Volume 11.05. Pp. 333-350.

Barron, M.G. 1990. Bioconcentration: Will water-borne organic chemicals accumulate in aquatic animals? *Environ. Sci. Technol.* 24:1612-1618.

Burkhard, L.P. and M.T. Lukasewycz. 2000. Some bioaccumulation factors and biota-sediment accumulation factors for polycyclic aromatic hydrocarbons in lake trout. *Environ. Toxicol. Chem*. 19:1427-1429.

Carr, K.H., G.T. Coyle and R.A. Kimerle. 1997. Bioconcentration of [^{14}C]butyl benzyl phthalate in bluegill sunfish (*Lepomis Macrochirus*). *Environ. Toxicol. Chem.* 16:2200-2203.

Connell, D.W. 1988. Bioaccumulation behavior of persistent organic chemicals with aquatic organisms. *Rev. Environ. Contam. Toxicol*. 101:117-159.

Connolly, J.P. and C.G. Pedersen. 1988. A thermodynamic-based evaluation of organic chemical accumulation in aquatic organisms. *Environ. Sci. Technol.* 22:99-103.

Cook, P.M. and L.P. Burkhard. 1998. Development of bioaccumulation factors for protection of fish and wildlife in the Great Lakes. In: *National Sediment Bioaccumulation Conference Proceedings*. U.S. Environmental Protection Agency, Office of Water. Washington, DC. EPA 823-R-002.

de Wolf, W., J.H.M. de Bruijn, W. Seinen and J.L.M. Hermans. 1992. Influence of biotransformation on the relationship between bioconcentration factors and octanol-water partition coefficients. *Environ. Sci. Technol.* 26:1197-1201.

Fisk, A.T., R.J. Norstrom, C.C. Cymbalisty and D.C.B. Muir. 1998. Dietary accumulation and depuration of hydrophobic organochlorines: Bioaccumulation parameters and their relationship with the octanol/water partition coefficient. *Environ. Toxicol. Chem.* 17: 951-961.

Gobas, F.A.P.C., J.R. McCorquodale and G.D. Haffner. 1993. Intestinal absorption and biomagnification of organochlorines. *Environ. Toxicol. Chem.* 12:567-576.

Gobas, F.A.P.C. 1993. A model for predicting the bioaccumulation of hydrophobic organic chemicals in aquatic food-webs: application to Lake Ontario. *Ecol. Mod.* 69:1-17

Hudson, R.J.M., A.S. Gherini, C.J. Watras and D.B. Porcella. 1994. Modeling the biogeochemical cycle of mercury in lakes: the Mercury Cycling Model (MCM) and its application to the MTL Study Lakes, In: C.J. Watras and J.W. Huckabee (eds.), *Mercury Pollution: Integration and Synthesis.* Lewis Publishers. Boca Raton, FL. Pp. 473-523.

Isnard, P. and S. Lambert. 1988. Estimating bioconcentration partition coefficients and aqueous solubility. *Chemosphere* 17:21-34.

Jafvert, C.T. 1990. Sorption of organic acid compounds to sediments: initial model development. *Environ. Toxicol. Chem.* 9:1259-1268.

Jafvert, C.T., J.C. Westall, E. Grieder and R.P. Schwarzenbach. 1990. Distribution of hydrophobic ionogenic organic compounds between octanol and water: Organic acids. *Environ. Sci. Technol*. 24:1795-1803.

James, M.O. 1989. Biotransformation and disposition of PAH in aquatic invertebrates, In: U. Varanasi (ed.). *Metabolism of Polycyclic Aromatic Hydrocarbons in the Aquatic Environment*. CRC Press, Inc. Boca Raton, FL. Pp. 69-92.

Mackay, D. 1982. Correlation of bioconcentration factors. *Environ. Sci. Technol.* 16:274-278.

Miranda, C.L., M.C. Henderson, and D.R. Buhler. 1998. Evaluation of chemicals as inhibitors of trout cytochrome p450s. *Toxicol. Appl. Pharmacol*. 148:327-244.

Niimi, A.J. 1985. Use of laboratory studies in assessing the behavior of contaminants in fish inhabiting natural ecosystems. *Wat. Poll. Res. J. Can.* 20:79-88.

Oliver, B.G. and A.J. Niimi. 1983. Bioconcentration of chlorobenzenes from water by rainbow trout: Correlations with partition coefficients and environmental residues. *Environ. Sci. Technol.* 17:287-291.

Oliver, B.G. and A.J. Niimi. 1988. Trophodynamic analysis of polychlorinated biphenyl congeners and other chlorinated hydrocarbons in the Lake Ontario ecosystem. *Environ. Sci. Technol*. 22:388-397.

Randall, R.C., H. Lee II, R.J. Ozretich, J.L. Lake and R.J. Pruell. 1991. Evaluation of Selected Lipid Methods for Normalizing Pollutant Bioaccumulation. *Environ. Toxicol. Chem.* 10: 1431-1436.

Randall, R.C., D.R. Young, H. Lee, and S.F. Echols. 1998. Lipid methodology and pollutant normalization relationships for neutral nonpolar organic pollutants. *Environ. Toxicol. Chem*. 17:788-791.

Russell, R.W., F.A.P.C. Gobas, and G.D. Haffner. 1999. Role of chemical and ecological factors in trophic transfer of organic chemicals in aquatic food webs. *Environ. Toxicol. Chem*. 18:1250-1257.

Schwarzenbach, R.P., P.M. Gschwend, and D.M. Imboden. 1993. *Environmental Organic Chemistry*. John Wiley and Sons, Inc. New York, NY.

Spacie, L.L. 1994. Interactions of organic pollutants with inorganic solid phases: are they important to bioavailability? In: *Bioavailability: Physical, Chemical and Biological Interactions.* Hamelink, J.L., Landrum, P.F., Bergman, H.L., and W.H. Benson (eds.). Proceedings of the Thirteenth Pellston Workshop, Pellston, MI. August 17-22, 1992. SETAC Special Publication Series. CRC Press, Inc. Boca Raton, FL. Pp. 73-82.

Suffet, I.H., C.T. Jafvert, J. Kukkonen, M.R. Servos, A. Spacie, L.L. Williams, and J.A. Noblet. 1994. Synopsis of discussion sessions: influence of particulate and dissolved material on the bioavailability of organic compounds, In: *Bioavailability: Physical, Chemical and*

Biological Interactions. Hamelink, J.L., Landrum, P.F., Bergman, H.L., and W.H. Benson (Eds.). Proceedings of the Thirteenth Pellston Workshop, Pellston, MI. August 17-22, 1992. SETAC Special Publication Series. CRC Press, Inc. Boca Raton, FL. Pp. 93-108.

Swackhamer, D.L. and R.A. Hites. 1988. Occurrence and bioaccumulation of organochlorine compounds in fishes from Siskiwit Lake, Isle Royale, Lake Superior. *Environ. Sci. Technol.* 22:543-548.

Thomann, R.V. 1989. Bioaccumulation model of organic chemical distribution in aquatic food chains. *Environ. Sci. Technol*. 23:699-707.

USEPA (U.S. Environmental Protection Agency). 1980. Appendix C–Guidelines and methodology used in the preparation of health effect assessment chapters of the consent decree water criteria documents. *Federal Register* 45:79347-79357. November 28.

USEPA (U.S. Environmental Protection Agency). 1991. *Technical Support Document for Water Quality-Based Toxics Control.* Office of Water. Washington, DC. EPA/505/2-90/001.

USEPA (U.S. Environmental Protection Agency). 1993. Assessment and control of bioconcentratable contaminants in surface water. *Federal Register* 56:13150.

USEPA (U.S. Environmental Protection Agency). 1995a. Final water quality guidance for the Great Lakes system; Final Rule. *Federal Register* 60:15366-15425. March 23.

USEPA (U.S. Environmental Protection Agency). 1995b. *Great Lakes Water Quality Initiative Technical Support Document for the Procedure to Determine Bioaccumulation Factors*. Office of Water. Washington, DC. EPA/820/B-95/005.

USEPA (U.S. Environmental Protection Agency). 1996. *Ecological Effects Test Guidelines. OPPTS 850.1730 Fish BCF. Public Draft.* Office of Prevention, Pesticides and Toxic Substances. Washington, DC. EPA/712/C-96/129. April.

USEPA (U.S. Environmental Protection Agency). 1997. Revisions to the polychlorinated biphenyl criteria for human health and wildlife for the water quality guidance for the Great Lakes system; Final Rule. *Federal Register* 62:11723-11731. March 12.

USEPA (U.S. Environmental Protection Agency). 2000a. *Trophic Level and Exposure Analyses for Selected Piscivorous Birds and Mammals. Volume I: Analyses of Species for the Great Lakes. Draft.* Office of Water. Washington, DC. August.

USEPA (U.S. Environmental Protection Agency). 2000b. *Trophic Level and Exposure Analyses for Selected Piscivorous Birds and Mammals. Volume II: Analyses of Species in the Conterminous United States. Draft.* Office of Water. Washington, DC. August.

USEPA (U.S. Environmental Protection Agency). 2000c. *Trophic Level and Exposure Analyses for Selected Piscivorous Birds and Mammals. Volume III: Appendices. Draft.* Office of Water. Washington, DC. August.

USEPA (U.S. Environmental Protection Agency). 2000d. *Technical Basis for the Derivation of Equilibrium Partitioning Sediment Guidelines (ESGs) for the Protection of Benthic Organisms: Nonionic Organics.* Office of Science and Technology, Office of Research and Development. Washington, DC. June Draft.

Veith, G.D., D.F.L. DeFoe and B.V. Bergstedt. 1979. Measuring and estimating the bioconcentration factor in fish. *J. Fish. Res. Board Can.* 36:1040-1045.